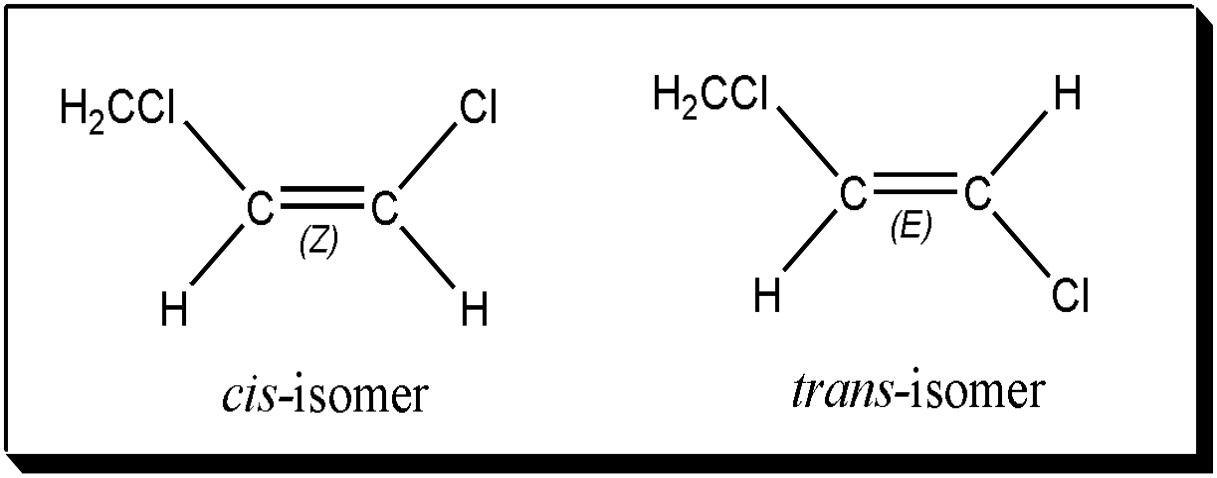
1,3-Dichloropropene

Risk Characterization Document

Inhalation Exposure to Workers, Occupational and Residential Bystanders and the General Public



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LIST OF ABBREVIATIONS

1,3-D	1,3-dichloropropene
3-CAA	3-chloroallyl alcohol
3-CPA	3-chloracrylic acid
AAC	annual air concentration
ATSDR	American College of Governmental Industrial Hygienists
AUR	air unit risk
AUC	air unit concentration
BMC	benchmark concentration
BMCL	95% lower bound on the benchmark concentration
CHO	Chinese hamster ovary (cells)
CDMS	California Data Management Systems
DPR	Department of Pesticide Regulation
ENEL	estimated no effect level
FIFRA	Federal Insecticide, Fungicide and Rodenticide Act
gd	gestation day
HEC	human equivalent concentration
LAC	lifetime air concentration
LC_{50}	air concentration resulting in death of half of the exposed animals
LOEL	lowest observed effect level
LRT	lower respiratory tract
NAC	N-acetyl cysteine
NOEL	no observed effect level
NPSH	non-protein sulfhydryl content
PCE	polychromatic erythrocytes
PISP	Pesticide Illness Surveillance Program (DPR)
ppb	parts per billion
ppm	parts per million
PUR	Pesticide Use Report
RCD	risk characterization document
RGDR	regional gas dose ratio
SAC	seasonal air concentration
STAC	short term air concentration
TIF	totally impervious film
TWA	time-weighted average
URT	upper respiratory tract

I. SUMMARY

1,3-Dichloropropene (1,3-D) is a fumigant used to control nematodes, insects and disease organisms in soil. It promotes crop growth by minimizing competition with soil pests. 1,3-D has major uses in California in fruit and nut trees, strawberries, grapes, carrots and a host of other food and non-food crops. It is commonly injected into soil on a preplant basis, after which it volatilizes and redissolves in the aqueous films that surround soil particles. It is also applied through post-plant drip irrigation. Regardless of the application method, volatilization creates the opportunity for off-site transport and subsequent human exposure.

1,3-D formulations consist primarily of *cis* (E) and *trans* (Z) isomers in approximately equal proportions, as well as lower concentrations of other dichloropropenes and stabilizers. Some formulations also contain chloropicrin, dibromoethane or methylisothiocyanate, though neither of the latter compounds are found in formulations currently registered in California.

While the mechanism of pesticidal action is unclear, 1,3-D may work by inactivating vital enzymes through formation of covalent bonds by nucleophilic displacement. In mammals, including humans, it is an irritant, asphyxiant and sensitizer, though again, precise mechanisms of action are not completely understood. The following human health assessment concentrates exclusively on risks arising from inhalation exposure projected to occur in California under occupational, bystander and ambient scenarios.

Illness and injury reports

In California between 1982-1990, 51 cases were reported to the California Department of Pesticide Regulation's Pesticide Illness Surveillance Program (PISP). Of these 51 cases, the health effects attributed to exposure to 1,3-D alone, or in combination with other pesticides, were rated as definite (33 cases), probable (9 cases) or possible (9 cases), The health effects involved were systemic (16 cases), eye (14), skin (18), and combined eye-skin effects (3). From 1990 to 1995, 1,3-D use in California was suspended. As a result, from 1990 to 1997, there were no reports of illnesses associated with 1,3-D applications. In 1998, PISP identified one possible case involving 1,3-D alone with no other cases appearing until 2002. Then, in the 10 years from 2002 to 2011, the PISP identified 17 exposure episodes that gave rise to 71 cases associated with 1,3-D either alone or in combination with chloropicrin . Those 71 cases were classified as 1 definite, 54 probable and 16 possible.

Of the 72 recently reported cases (*i.e.*, between 1998 and 2011), there were 5 cases with 1,3-D used alone between 1998 and 2011. Four of these cases exhibited respiratory symptoms. In 2007, 1 episode involving 3 cases was reported. The remaining 67 cases involved both 1,3-D and chloropicrin. Most of the cases involving 1,3-D and chloropicrin show dominance of eye effects, suggesting that the reported eye symptoms may be due to the chloropicrin.

Environmental fate

Soil. The half-life ($t_{1/2}$) of 1,3-D in soil varies with soil type, temperature and microbial content, but can range as high as 69 days. Interestingly, the soil $t_{1/2}$ is lower under anaerobic conditions, ranging between 2.4 and 9.1 days. Adsorption of 1,3-D to soil is stronger in the vapor phase, which dominates at normal soil temperatures, than in water. Diffusion (as opposed to leaching) is the primary mode of soil mobility. Hydrolysis in soil is influenced by moisture content and organic matter, and follows pseudo first-order kinetics to yield chloroallyl alcohol and other degradates. While runoff rates are low due to high soil persistence and high hydrolysis rates, 25-56% of applied 1,3-D will volatilize within 2 weeks.

Water. The volatilization $t_{1/2}$ of 1,3-D from surface water was determined to be ~4 hours based on modeling of a river, though its maximum estimated $t_{1/2}$ is 50 hours when the model is of a pond and includes adsorption to particulates . Hydrolysis in water occurs over periods of days to weeks. Photolysis occurs over a much longer period, so is not a significant factor in 1,3-D degradation. 1,3-D is unlikely to be a groundwater contaminant.

Air. As noted, 1,3-D readily volatilizes into air---about 40% within 2-3 weeks---thus creating the potential for inhalation exposure. The tropospheric $t_{1/2}$ of the *trans* and *cis* isomers is 30 and 50 hours, respectively. Decomposition is aided by atmospheric particulates, light intensity and NO_2 concentration. In addition, airborne 1,3-D is degraded by hydroxyl radicals ($t_{1/2}$ = 7 hours and 12 hours for the *trans* and *cis* isomers, respectively) and by ozone ($t_{1/2}$ = 12-52 days). 1,3-D is also photooxidized to 3-chloropropionyl chloride and subsequently hydrolyzed to 3-chloropropionic acid, which is removed by rainfall.

Vegetation. 1,3-D applied to soils usually dissipates before planting, which, in addition to its rapid metabolism in cases when it *is* absorbed, makes it unlikely that residues will be found in food crops. The measured Log K_{OW} of 1.82 does not indicate a potential for bioaccumulation in aquatic and terrestrial food chains.

Pharmacokinetics

The inhalation and oral absorption of 1,3-D is evident in the detection of the parent compound and metabolites in urine and tissues, and by the observed systemic toxicity.

Inhalation exposure in humans. A study in which human volunteers were exposed by inhalation for 6 hours to 1 ppm *cis/trans* 1,3-D showed that respiratory uptake was ~80% for both isomers. Initial phase half-lives for urinary excretion of *cis* and *trans* N-acetyl cysteines (major conjugation products of 1,3-D) were 4.2 and 3.2 hours, respectively. Terminal phase half-lives were 12.3 and 17.1 hours. This study showed rapid and near-complete (~80%) uptake through the lung and subsequent metabolism to excretable compounds in humans.

Inhalation exposure in rats. A study in which rats were exposed under nose-only conditions to 1,3-D at 30, 90, 300 or 900 ppm for 3 hours generated estimated absorbed doses of 82% at 30

ppm and 62-65% at 90-900 ppm. Blood levels were constant at 30-90 ppm during exposure, falling rapidly after the end of exposure (especially for *cis*-1,3-D). At 300 and 900 ppm, blood levels rose from hour to hour during exposure. The *trans* isomer tended to have higher blood concentrations than the *cis* isomer, probably due in part to slower metabolism or excretion. Using special surgical procedures to isolate the lower and upper respiratory tracts, it was determined that ~50% of the available 1,3-D was absorbed in the lower tract and 11-16% in the upper tract. Respiratory frequency decreased at 300 and 900 ppm, though tidal volume stayed the same. The lowered frequency and percent absorption led to >50% reduction in 1,3-D uptake at 900 ppm. The presence of separate anatomical areas in the respiratory tract for absorption of 1,3-D is toxicologically significant because it identifies both different areas for potential portal of entry effects and systemic toxicities that might be absorption-site dependent.

Oral exposure in rats. Rats were exposed to 5 mg/kg 1,3-D (*cis* and *trans* isomers) by gavage in corn oil for 14 consecutive days, after which they were fasted for 8 hours prior to dosing with 5 mg/kg uniformly labeled ^{14}C-1,3-D and sacrificed 48 hours later. Approximately 62-65% of the administered dose was found in urine, 26% in exhaled air (as CO_2), 5% in feces, and 4-6% in tissues and carcass, with no observed sex differences. Disposition in non-pre-treated rats was similar. At 48 hours after dosing of pre-treated rats, the highest concentrations of label (about 1 µg equivalent/g tissue) were in bladder and forestomach, and the lowest concentrations were in brain and fat (less than 0.1 µg equivalent/g tissue). Urinary excretion of the *cis/trans* N-acetyl cysteine conjugates comprised 26-28% of administered dose. The combined close-eluting sulfoxide and sulfone residues (*i.e.*, oxidation products of the NAC conjugates) comprised about 14% of administered dose. No other components were characterized. Most excretion occurred within the first 12 hours. The investigators proposed a reaction scheme in which 1,3-D is either conjugated with glutathione, ultimately converted through peptidase action to the cysteine conjugate and excreted through the urine or converted to acetyl CoA and CO_2 through the intermediates 3-chloallyl alcohol and 3-chloroacrolein.

Two further studies from the open literature examined the fate of orally administered 1,3-D in rodents, with emphasis on the formation and potential effects of specific metabolites. The general conclusion from these studies was that while metabolism of 1,3-D leads mostly to relatively innocuous products. However, a small but toxicologically significant portion of the absorbed dose is metabolized to reactive species of toxicologic concern, including potentially genotoxic epoxides and their derivatives.

Acute toxicity

Acute or short-term inhalation exposure to high concentrations of 1,3-D results in upper respiratory symptoms in humans, including chest tightness, irritated and watery eyes, dizziness and runny nose. Laboratory rats exposed for 4 hours to 1,3-D vapors at doses of 300 ppm and higher exhibited labored, slow or exaggerated breathing; liver and lung congestion; lung edema; hydrothorax; corneal opacity; closed eyelids; visceral congestion; hunched posture; restlessness;

pawing behavior; and body weight losses or body weight gain reductions. Death was often observed above 600 ppm.

Data on clinical or pathologic signs were not adequate in strictly acute inhalation studies to set thresholds for acute toxicity, as the high dose ranges used in those studies were designed to determine LC_{50}s. Consequently, toxicologic studies not limited to a single day treatment but reporting findings shortly after the onset of exposure (usually up to 7 consecutive days) were considered for identifying acute NOELs. Developmental toxicity studies were of particular interest because the reported endpoints can potentially result from a single or short-term exposure.

Nine inhalation subchronic, chronic and developmental toxicity studies reported effects occurring at early time points. The most common and sensitive effects in these studies were reductions in body weight and/or body weight gain, observed in rats, mice and rabbits. Virtually no other clinical signs or pathologies were noted at the dose ranges employed. The body weight effects, which were likely generalized expressions of animal stress, occurred early in the treatment period (*i.e.*, within 1-13 days) and continued after repeated exposures at the higher end of the concentration ranges employed. Comparable concentrations of 1,3-D in different species elicited comparable decrements in body weight or weight gain.

Benchmark concentration (BMC) analysis was undertaken to determine the critical acute endpoint value in animals. In this approach, the BMC is the lower 95% confidence limit of the effective dose (BMCL) required to induce a particular response. A benchmark response (BMR) of 1 standard deviation (SD, 1σ) for unexposed animals was selected to analyze body weight effects. The data in 6 of the 9 studies (five in rats, one in mice) could be modeled with the algorithms available in the Benchmark Dose Software. The estimated acute inhalation $BMCL_{1\sigma}$ values ranged between 40 and 66 ppm, with the lowest value of 40 ppm resulting from analysis of body weight decrements in female mice occurring during the first week of the 2-year chronic mouse study. However, the 13-week rat study established a BMCL of 49 ppm based on weight decrements first measured in males at 3 days. Because this time period most closely approximated an acute exposure regimen, **49 ppm** was selected as the critical endpoint value used to evaluate risks arising from acute / short-term exposures. The Regional Gas Dose Ratio (RGDR) approach was used to adjust the dose in the animal inhalation experiments to Human Equivalent Concentrations (HEC). This adjustment takes into account physiological and anatomical differences between humans and animals. Application of the appropriate RGDR scalar to the BMCL of 49 ppm for body weight decreases in rats resulted in HECs of **33 and 11 ppm** for occupational and non-occupational exposure scenarios. These values were used to estimate acute / short-term risk from 1,3 D exposure.

Subchronic toxicity

The critical inhalation $BMCL_{10}$ in laboratory animals for the evaluation of seasonal exposure risks was **16 ppm**. This was based on the appearance of hyperplasia of the nasal respiratory

epithelium in rats at 30 ppm after 13 weeks of daily exposure (5 days/week, 6 hr/day). Application of the RGDR scalar resulted in occupational and non-occupational HECs of 0.90 and 0.30 ppm. These values were used to estimate subchronic / seasonal risk.

Chronic toxicity

The critical inhalation $BMCL_{10}$ for the evaluation of chronic exposure scenarios was **6 ppm**. This was based on the 2-year inhalation study in the mouse, which showed hyperplasia of the nasal respiratory epithelium in females, hyperplasia and hypertrophy of the urinary bladder transitional epithelium in males and females, and roughened, irregular opaque urinary bladder surface in females at the LOEL dose of 20 ppm. Those signs increased in incidence and severity at the high dose of 60 ppm. Application of the RGDR scalar resulted in occupational and non-occupational HECs of 0.59 and 0.20 ppm. These values were used to estimate chronic / annual risk.

Oncogenicity

In 2007, the USEPA classified 1,3-D as "likely to be carcinogenic to humans" based on animal studies. The International Agency for Research on Cancer (IARC) grouped 1,3-dichloropropene as a group 2B carcinogen ("possibly carcinogenic to humans").

Male mice exposed to 1,3-D by the inhalation route for 2 years exhibited a statistically elevated incidence of bronchioloalveolar adenomas at a nominal air concentration of 60 ppm (22/50: 44%). The historical control incidence range for 7 studies using the same strain of mice and conducted in the same laboratory was 7 - 32%. The incidence rate at 20 ppm (13/49: 27%) was higher than concurrent controls (9/49: 18%) and was at the high end of the historical control range. In view of the apparent dose dependence and the evidence for genotoxicity, the linearized multistage cancer model (BMCS version 2.6) was used to characterize the dose response. The multistage model is considered standard for cancer bioassay modeling when there is no evidence for a threshold model and when the data are amenable to dose-response modeling, as is the case here. It is also used when there is evidence for genotoxicity, also the case for 1,3-D. A benchmark response (BMR) of 10% "extra risk" was chosen to determine the slope potency, referred to in this document as the air unit risk (AUR). Application of the appropriate RGDR scalar to the dose levels used in the 2-year study, followed by BMC modeling of the incidence rates, generated AUCs (Air Unit Concentration) of 0.0059 ppm^{-1} and 0.018 ppm^{-1} for occupational and non-occupational exposure scenarios, respectively, assuming a portal of entry mode of action. However, after consideration of all of the available data, we concluded that a systemic mode of action could not be excluded, necessitating determination of parallel *systemic* AUCs: 0.02 ppm^{-1} and 0.062 ppm^{-1} for occupational and non-occupational exposures. Because non-occupational cancer risk was calculated only for ambient scenarios, the non-occupational AUCs were converted to conventional cancer potency values: 0.000014 $(\mu g/kg/day)^{-1}$ and 0.000048 $(\mu g/kg/day)^{-1}$ for portal of entry and systemic modes of action. This was done because

the requisite morphometric information---in particular, respiratory tract surface areas for children and immature mice---was not available. Consequently, these age specific air unit risk values were substituted by potency values that took into account relative breathing rates normalized to body weight for immature subpopulations.

Reproductive and developmental toxicity

There was little indication from the reproductive and developmental inhalation toxicity studies that 1,3-D poses a health risk to humans with respect to these parameters.

Exposure estimation

1,3-D is present in 17 soil fumigation products currently available in California. The fumigants can be applied via shank or hand-wand injection, and drip irrigation. Thirteen of the 17 products also contain chloropicrin. 1,3-D breathing-zone air concentrations were estimated for worker and bystander exposure scenarios. The worker scenarios included handlers applying 1,3-D via the use of shallow shank injection, deep shank injection, drip irrigation, or the injection auger. The shank and drip applicators treat soils with or without the use of a tarpaulin. In addition to applicators, 1,3-D breathing-zone air concentrations were estimated for handlers removing the tarp(s) from the fumigated field. 1,3-D breathing-zone air concentrations were also estimated for the worker who loaded the fumigant for application, and the reentry worker that entered the treated field following the restricted entry interval. The bystander scenarios considered include the occupational bystander which could be adjacent to a field undergoing fumigation via shallow or deep shank without the use of a tarp, or via drip irrigation with the use of a tarp. In addition, 1,3-D breathing-zone air concentrations for the residential bystander located 100 feet from the edge of the treated field were estimated. The 100-foot buffer zone is mandated by CA permit conditions for any occupied structures (e.g., residences, schools), and, as a result, was applied for the residential bystander when estimating exposure due to a nearby application. However, an occupational bystander could potentially be a field worker adjacent to the field being treated. There's no language in the CA permit conditions and certain product labels addressing this scenario. Hence, for the occupational bystander, the buffer zone was not incorporated into the exposure assessment. The estimates for the residential bystander were classified according to the potential source of 1,3-D (i.e., from the nearby treated field or from ambient air).

The 1,3-D breathing-zone air concentrations were estimated for short-term, seasonal, annual, and lifetime exposure periods. The short-term exposures consist of 8 hours a day for up to one week for the occupational scenarios, and 24-hrs for up to one week for the residential bystander. Seasonal exposure consists of the daily breathing-zone 1,3-D air concentration for the total use season which is defined as the months of the year where the number of pounds of 1,3-D applied is equal to or greater than 5 percent of the annual total. Annual exposure represents the daily breathing-zone air concentration of 1,3-D over the course of the entire year. Lifetime exposure represents the daily air concentration to which a worker or bystander is exposed over a lifetime.

The estimated breathing-zone 1,3-D short-term air concentrations for the workers ranged from 0.037 ppm (8-hr TWA) to 33 ppm (8-hr TWA) with the lowest and highest concentrations for the reentry worker and tarp remover scenarios, respectively. The occupational bystander short-term breathing-zone air concentrations were simulated for 8 hours TWA for different application methods and field sizes: shallow shank application (80 acres and without a tarp), deep shank application (80 acres and without a tarp), and drip application (40 acres and with a tarp). The simulated short-term air concentrations for these scenarios are 2, 0.6, 1.1 ppm, respectively. The estimated 1,3-D residential bystander breathing-zone short-term air concentrations due to the fumigation of a nearby field, were simulated for 24 hours TWA for different application methods and field sizes: shallow shank application (80 acres and without a tarp), deep shank application (80 acres and without a tarp), and drip application (40 acres and with a tarp). The simulated short-term air concentrations for these scenarios are 0.5508, 0.1432, and 0.1800 ppm, respectively. Since a large percentage of 1,3-D is used for treating soil used for planting trees and vines, a simulation was carried out specifically for these crops for residential bystander exposure. Only short-term exposure is anticipated, so only the short-term air concentration (i.e., 0.0918 ppm for 24 hours TWA), was simulated for this scenario. The other potential source of short-term exposure anticipated for the residential bystander is 1,3-D in ambient air. This was estimated via 72-hour air concentration measurements taken throughout the year within Merced County. The highest of these measured 72-hr TWA air concentrations, used as the short-term air concentration, is 0.0813 ppm.

The estimated breathing-zone 1,3-D seasonal air concentrations for workers ranged from 0.0045 ppm (8-hr TWA) to 8.3 ppm (8-hr TWA) with the lowest and highest concentrations for the occupational bystander and tarp remover scenarios, respectively. The estimated 1,3-D residential bystander breathing-zone seasonal air concentrations (24 hr TWA) were obtained from simulated air concentrations for the previously described scenarios. These air concentrations are 0.0173, 0.0135, and 0.0050 ppm for the shallow shank, deep shank, and drip application scenarios, respectively. The estimated seasonal 1,3-D air concentration in ambient air for the residential bystander was made equal to 0.0045 ppm, the mean of the air concentrations measured during the continuous 1,3-D use season in Merced County.

The estimated breathing-zone 1,3-D annual air concentrations for workers ranged from 0.00062 ppm (8-hr TWA) to 5.2 ppm (8-hr TWA) with the lowest and highest concentrations for the occupational bystander and tarp remover scenarios, respectively. For the residential bystander, the 1,3-D annual air concentrations due to the fumigation of a nearby field were not estimated because they were considered indistinguishable from the 1,3-D concentrations in ambient air. The estimated annual 1,3-D air concentration in ambient air for the residential bystander was made equal to 0.0002 ppm, the median of 129,600 computer-simulated ambient air concentration values in Merced County.

The estimated breathing-zone 1,3-D lifetime air concentrations for workers ranged from 0.0003 ppm (8-hr TWA) to 2.8 ppm (8-hr TWA) with the lowest and highest concentrations for the occupational bystander and tarp remover scenarios, respectively. For the residential bystander, the 1,3-D lifetime-air concentrations due to the fumigation of a nearby field were not estimated because they were considered indistinguishable from the 1,3-D concentrations in ambient air. Lifetime ambient 1,3-D exposures of residential bystanders were generated using simulated air concentrations and two computer models: MCABLE and HEE5CB. The 95th percentile ambient exposure estimates varied depending on model, gender and assumptions regarding residence time in the affected area, from as low as 0.1644 μg/kg/day to as high as 0.8396 μg/kg/day.

Risk analysis

Non-oncogenic risk estimates. The potential for non-oncogenic health effects resulting from exposure to 1,3-D was expressed as the margin of exposure (MOE), which was calculated as follows:

Margin of Exposure (MOE) = HEC (in ppm) / Exposure dose (in ppm)

As this assessment was focused on risks arising from inhalation exposure to 1,3-D vapor, both the HEC and the exposure values were expressed as air concentrations (ppm) rather than as internal doses (mg/kg).

For adults under occupational or non-occupational exposure conditions, MOEs of 30 were considered adequate to protect human health. This "target MOE" was the product of an uncertainty factor of 3, to account for pharmacodynamic differences between laboratory animals and humans, and 10 to account for an assumed 10-fold range of sensitivity within the human population. For children, who are presumably exposed only under non-occupational scenarios, the target MOEs was 100. The extra ~3-fold factor was due to database uncertainty arising because no toxicity studies were conducted on young animals. Consequently, we had no way of assessing the possibility that infants and children might be more susceptible to the toxic effects of 1,3-D. In addition, the lack of default surface area values for infants and children precluded RGDR-based calculations for those demographics.

MOEs for acute / short-term occupational scenarios were calculated by applying the critical occupational HEC of 33 ppm to the acute / short-term occupational exposure estimates. The highest risk acute / short-term risk occupation was tarp remover, which showed MOEs of 1 for various application types. Two additional occupational scenarios resulted in MOEs below the target of 30--- applicator (shallow shank without tarp; MOE = 17) and applicator (injection auger; MOE = 28).

Acute / short-term non-occupational MOEs were calculated by applying the critical HEC value of 11 ppm to the short-term exposure estimates. The lowest MOE of 20 was determined for a resident / bystander at the edge of a buffer zone for a shallow shank application. This value was

below both the adult and child target MOEs of 30 and 100, respectively. No other MOEs were below 30. However, two of the remaining scenarios---near an application site at the edge of a buffer zone both for deep shank application and drip application---showed MOEs below 100 (MOE = 96 and 61, respectively). Two other scenarios---at the edge of the buffer zone for a tree and vine application and ambient---showed MOEs greater than 100.

MOEs for subchronic / seasonal occupational scenarios were calculated by applying the critical occupational HEC of 0.90 ppm to the seasonal occupational exposure estimates. The highest seasonal risk occupation was tarp remover, which showed MOEs of less than 1 for three different application types. Several additional occupational scenarios resulted in MOEs below the target MOE of 30, including applicator (shallow shank without tarp; MOE = 28), applicator (shallow shank with tarp; MOE = 9), applicator (deep shank without tarp; MOE = 13), applicator (deep shank with tarp; MOE = 4), applicator (drip without tarp; MOE = 23), loader (shallow shank; MOE = 15), loader (deep shank; MOE = 7) and reentry worker (deep shank; MOE = 28).

MOEs for subchronic / seasonal risk for non-occupational scenarios were calculated by applying the critical HEC value of 0.30 ppm to the seasonal exposure estimates. The lowest MOE of 17 was determined for exposure at the edge of a buffer zone for a shallow shank application. This value was below both the adult and child target MOEs of 30 and 100, respectively. One additional seasonal exposure scenario showed a sub-30 MOE---near an application site, edge of buffer zone, deep shank (MOE = 22). Exposure near an application site, edge of buffer zone, drip gave an MOE of 60, indicating a seasonal health risk to children. This was also true for ambient exposure, with its seasonal MOE of 67.

MOEs for chronic / annual occupational scenarios were calculated by applying the critical occupational HEC of 0.59 ppm to the annual occupational exposure estimates. The highest annual occupational risk was tarp remover, which showed MOEs of less than 1 for three different application types. Several additional occupational scenarios resulted in MOEs below the target MOE of 30, including applicator (shallow shank with tarp; MOE = 18), applicator (deep shank without tarp; MOE = 14), applicator (deep shank with tarp; MOE = 4), loader (deep shank; MOE = 7) and reentry worker (deep shank; MOE = 25).

An MOE of 1000 was calculated using the critical non-occupational HED of 0.20 ppm for chronic / annual ambient risk. No other non-occupational exposure scenarios were anticipated.

Oncogenic risk estimates. For oncogenic effects, risk estimates less than the negligible risk standard of 10^{-6} were considered sufficient to protect human health. These estimates were based on the appearance of bronchioloalveolar adenomas in males in a 2-year mouse inhalation study. Because the evidence did not overwhelmingly favor either a portal of entry or a systemic mode of oncogenic action, we opted to express cancer risk for both routes (though we felt that the evidence tilted to the portal of entry scenario). Hence oncogenic risk was calculated using the

upper confidence limit (UCL) slope values---referred to as the air unit risk---of 0.0059 ppm^{-1} (portal of entry) and 0.020 ppm (systemic) for occupational scenarios, and 0.000014 [µg/kg/day]-1 (portal of entry) and 0.000048 [µg/kg/day]-1 for ambient lifetime exposure scenarios. These values were multiplied by the relevant lifetime exposures for the scenarios characterized in this analysis.

All of the occupational and ambient lifetime exposure scenarios showed oncogenic risk values that were above the negligible oncogenic risk standard of $1x10^{-6}$, regardless of assumed mode of action. Occupational cancer risk values for a portal of entry mode of action ranged between $1.9x10^{-6}$ (occupational bystander near an application site, 3 scenarios) and $1.7x10^{-2}$ (tarp remover, deep shank); for a systemic mode of action they ranged between $6.6x10^{-6}$ (occupational bystander near an application site, 3 scenarios) and $5.6x10^{-2}$ (tarp remover, deep shank) . Ambient cancer risks ranged between $2.30x10^{-6}$ (portal of entry, Mcable, 30-yr fixed, female) and $40.44x10^{-6}$ (systemic, HEE5CB, birth to age 70, low mobility). All of the ambient scenarios (lifetime) showed oncogenic risk values that were above the negligible risk standard of 1×10^{-6}. Depending on the residency-mobility assumptions employed, actual risk values ranged between $(2.30\text{-}4.66) \times 10^{-6}$ for the portal-of-entry effect and $(7.91\text{-}16.02) \times 10^{-6}$ for the systemic effect with MCABLE; $(4.75\text{-}11.75) \times 10^{-6}$ for the portal-of-entry effect and $(16.34\text{-}40.44) \times 10^{-6}$ for the systemic effect with HEE5CB.

All of the work tasks examined showed oncogenic risk values that exceeded the negligible oncogenic risk standard of $1x10^{-6}$. Actual oncogenic risk values calculated using the cancer potency factor assumptions of portal-of-entry and systemic effects ranged from $(4.46\text{-}15.04) \times 10^{-6}$ (i.e., occupational bystanders) to $(10321.94\text{-}34780.45) \times 10^{-6}$ (tarp remover [deep shank]).

Target MOEs and calculated MOEs for all exposure scenarios appear in Summary Table I. Oncogenic risk values for all occupational exposure scenarios appear in Summary Table II. Oncogenic risk values for ambient exposure scenarios appear in Summary Table III.

Summary Table I. Target MOEs and calculated MOEs for non-occupational and occupational 1,3-D exposure scenarios

Exposure scenario	Target MOE	Calculated MOE		
		Acute / short term	Seasonal	Annual
Occupational scenarios				
Applicator				
■ shallow shank w/o tarp	30 (adult)	122	28	61
■ shallow shank w/ tarp		39	9	18
■ deep shank w/o tarp		122	13	14
■ deep shank w/ tarp		39	4	4
■ drip w/o tarp		118	23	45
■ drip w/ tarp		143	50	98
■ injection auger		28	n/a	n/a
Loader				
■ shallow shank		47	15	31
■ deep shank		47	7	7
Tarp remover				
■ shallow shank		1	0.23	0.49
■ deep shank		1	0.11	0.11
■ drip		1	0.35	0.69
Reentry worker				
■ shallow shank		892	60	92
■ deep shank		892	28	25
■ drip		892	90	134
Occupational bystander				
■ shallow shank w/o tarp		17	750	952
■ deep shank w/o tarp		55	750	952
■ drip w/ tarp		30	750	952
Non-occupational scenarios				
Near application site, edge of buffer zone				
■ shallow shank	30 (adult); 100 (child)	20	17	n/a
■ deep shank		96	22	n/a
■ drip		61	60	n/a
■ tree & vine		120	n/a	n/a
Ambient		135	67	1000

Summary Table II. Oncogenic risk values for occupational 1,3-D exposure scenarios

Exposure scenario	Calculated oncogenic risk (negligible oncogenic risk standard = $1x10^{-6}$)	
Occupational scenarios		
	Portal of entry	**Systemic**
Applicator		
■ shallow shank w/o tarp	$3.2x10^{-5}$	$1.1x10^{-4}$
■ shallow shank w/ tarp	$1.0x10^{-4}$	$3.4x10^{-4}$
■ deep shank w/o tarp	$1.4x10^{-4}$	$4.6x10^{-4}$
■ deep shank w/ tarp	$4.3x10^{-4}$	$1.4x10^{-3}$
■ drip w/o tarp	$4.1x10^{-5}$	$1.4x10^{-4}$
■ drip w/ tarp	$1.9x10^{-5}$	$6.4x10^{-5}$
■ injection auger	n/a	n/a
Loader		
■ shallow shank	$5.9x10^{-4}$	$2.0x10^{-4}$
■ deep shank	$2.6x10^{-4}$	$8.8x10^{-4}$
Tarp remover		
■ shallow shank	$3.9x10^{-3}$	$1.3x10^{-2}$
■ deep shank	$1.7x10^{-2}$	$5.6x10^{-2}$
■ drip	$2.7x10^{-3}$	$9.2x10^{-3}$
Reentry worker		
■ shallow shank	$2.0x10^{-5}$	$6.8x10^{-5}$
■ deep shank	$7.1x10^{-5}$	$2.6x10^{-4}$
■ drip	$1.4x10^{-5}$	$4.8x10^{-5}$
Occupational bystander		
■ shallow shank w/o tarp	$1.9x10^{-6}$	$6.6x10^{-6}$
■ deep shank w/o tarp	$1.9x10^{-6}$	$6.6x10^{-6}$
■ drip w/ tarp	$1.9x10^{-6}$	$6.6x10^{-6}$

Summary Table III. Oncogenic risk values for ambient exposure scenarios

Ambient exposure scenarios – 95th percentile oncogenic risk				
	Portal of entry		Systemic	
	Male	Female	Male	Female
MCABLE				
With time away				
■ **Variable**	2.64×10^{-6}	2.49×10^{-6}	9.09×10^{-6}	8.56×10^{-6}
■ **30-yr fixed**	2.45×10^{-6}	2.30×10^{-6}	8.44×10^{-6}	7.91×10^{-6}
■ **50-yr fixed**	3.27×10^{-6}	3.04×10^{-6}	11.26×10^{-6}	10.44×10^{-6}
■ **70-yr fixed**	4.28×10^{-6}	3.97×10^{-6}	14.72×10^{-6}	13.64×10^{-6}
Without time away				
■ **Variable**	2.87×10^{-6}	2.69×10^{-6}	9.88×10^{-6}	9.23×10^{-6}
■ **30-yr fixed**	2.63×10^{-6}	2.49×10^{-6}	9.06×10^{-6}	8.57×10^{-6}
■ **50-yr fixed**	3.60×10^{-6}	3.31×10^{-6}	12.40×10^{-6}	11.39×10^{-6}
■ **70-yr fixed**	4.66×10^{-6}	4.27×10^{-6}	16.02×10^{-6}	14.68×10^{-6}
HEE5CB				
High mobility				
■ **Birth to age 30**	4.85×10^{-6}	4.75×10^{-6}	16.70×10^{-6}	16.34×10^{-6}
■ **Birth to age 50**	6.99×10^{-6}	6.76×10^{-6}	24.08×10^{-6}	23.25×10^{-6}
■ **Birth to age 70**	9.18×10^{-6}	8.77×10^{-6}	31.56×10^{-6}	30.17×10^{-6}
Intermediate mobility				
■ **Birth to age 30**	5.88×10^{-6}	6.97×10^{-6}	20.22×10^{-6}	23.99×10^{-6}
■ **Birth to age 50**	9.01×10^{-6}	8.16×10^{-6}	30.98×10^{-6}	28.08×10^{-6}
■ **Birth to age 70**	10.77×10^{-6}	10.6×10^{-6}	37.06×10^{-6}	35.45×10^{-6}
Low mobility				
■ **Birth to age 30**	6.25×10^{-6}	7.73×10^{-6}	21.51×10^{-6}	26.57×10^{-6}
■ **Birth to age 50**	10.43×10^{-6}	8.83×10^{-6}	35.89×10^{-6}	30.38×10^{-6}
■ **Birth to age 70**	11.75×10^{-6}	11.46×10^{-6}	40.44×10^{-6}	39.41×10^{-6}

Reference concentrations

Reference Concentrations (RfC: estimates of inhalation exposures to humans that are likely to be without appreciable risk of deleterious effects) for 1,3-D were generated for different age groups and exposure scenarios. These values were calculated by dividing the critical endpoint concentrations (expressed as human equivalent concentrations) by the uncertainty factors appropriate to the exposure scenarios evaluated. Uncertainty factors for 1,3-D were specified below in Table IV.4. Because the product of the uncertainty factors is equal to the target MOE, RfCs are simply the critical HEC divided by the target MOE.

RfCs for occupational scenarios (calculated only for adults) were 1100, 30 and 20 ppb for acute / short-term, seasonal and annual scenarios, respectively.

RfCs for non-occupational scenarios in adults were 367, 10 and 7 ppb for short-term, seasonal and annual scenarios, respectively. RfCs for non-occupational scenarios in children were 110, 3 and 2 ppb for those three exposure scenarios.

Conclusions

Acute / short term occupational MOEs (analyzed for adults only): Five of the 18 scenarios examined showed calculated MOEs lower than the target MOE of 30. Work tasks for these 5 scenarios included tarp removers (3 tasks), applicators (1 task), and occupational bystanders (1 task).

Subchronic / seasonal occupational MOEs: Seasonal occupational exposure scenarios showed 11 of 18 scenarios with calculated MOEs lower than the target MOE of 30. The low-MOE scenarios included applicators (5 tasks), loaders (2 tasks), tarp removers (3 tasks) and reentry workers (1 task).

Chronic / annual occupational MOEs: Annual occupational exposure scenarios showed 8 of 18 scenarios with calculated MOEs lower than the target MOE of 30. The low-MOE scenarios included applicators (3 tasks), loaders (1 task), tarp removers (3 tasks) and reentry workers (1 task).

Acute / short term resident / bystander (non-occupational) MOEs: In adults, 1 of the 4 scenarios showed a calculated MOE lower than the target MOE of 30, while in children 3 of the same 4 scenarios showed calculated MOEs lower than the target MOE of 100.

Seasonal resident / bystander (non-occupational) MOEs: In adults, 2 of the 3 scenarios showed a calculated MOE lower than target MOE of 30, while in children all three scenarios were lower than the target MOE of 100.

Annual resident / bystander (non-occupational) MOEs: Annual exposure to residential bystanders was not expected.

Ambient Air MOEs, acute / short term, subchronic / seasonal and chronic / annual: The acute / short term ambient MOE of 135 exceeded both the target MOEs of 30 for adults and 100 for children. The subchronic / seasonal ambient MOE of 67 exceeded the target MOE of 30 for adults, but did not exceed the target MOE of 100 for children. The chronic / annual ambient MOE of 1000 exceeded the target MOE of 30 for adults both the target MOEs of 30 for adults and 100 for children.

Oncogenic risk, occupational and ambient: Calculated oncogenic risk values were higher than the negligible oncogenic risk standard of $1x10^{-6}$ for every occupational and non-occupational scenario analyzed including all ambient scenarios, which were analyzed using computer simulations. This was the case regardless of the assumed oncogenic mode of action---portal of entry or systemic---of 1,3-D.

Risk appraisal – toxicology

Uncertainties regarding the toxicologic mode of action of 1,3-D lead to uncertainties in the resultant risk characterization. The most prominent of these toxicologic uncertainties included:

■ The use of body weight decrements to drive the acute/ short term risk evaluation. As this was the only toxicologic effect at low doses, a decision not to use body weight, but an estimated NOEL of 77.5 ppm based on clinical signs occurring at a near-lethal dose of 775 ppm would have resulted in 1.6-fold higher HECs and higher MOEs.

■ The assumption that the acute / short term body weight decrements resulted from systemic, as opposed to portal-of-entry, exposure. Had the body weight decrements been considered the result of interaction with extrathoracic (*eg*., nasal) passages, the HEC and resultant MOE would have been lower than the systemic HEC and MOE. On the other hand, had the body weight decrements been considered the result of interaction with tracheobronchial and pulmonary passages, the HEC and resultant MOE would have been higher.

■ The decision to reduce the 3x pharmacokinetic uncertainty factor to 1x because the RGDR approach was used in the acute / short term assessment. Retention of this factor would have resulted in target MOEs that were 3-fold higher than those used in this assessment.

■ The decision to base the chronic / annual assessment on nasal hyperplasia in the mouse necessitated the use of an extrathoracic mouse-to-human RGDR scalar of 0.198 to calculate HECs of 0.16 and 0.49 ppm for non-occupational and occupational scenarios, respectively. However, bladder effects were also noted in the critical study at the same air concentration, demonstrating that in addition to a portal of entry effect, 1,3-D also had systemic effects under

chronic conditions. Had the critical chronic value been based on bladder effects, the HEC and resultant MOEs would have been ~5-fold higher.

■ The decision to base the calculation of HECs for the subchronic / seasonal and the chronic / annual assessments on rodent-to-human extrathoracic RGDR scalars of 0.115 and 0.198. However, data from some chemicals suggest that the extrathoracic RGDR may be closer to 1 (*i.e.*, similar to the default systemic RGDR). If so, this would suggest that the critical seasonal and annual HECs may be underestimates by as much as 9- and 5-fold, respectively.

■ With the exception of genotoxicity, the effects of metabolites, degradates and impurities in the toxicity studies were not evaluated in this assessment.

Risk appraisal - exposure

In order to generate exposure estimates, certain approaches were taken or assumptions made which create uncertainty:

■ Surrogate data from chloropicrin exposure studies were used to estimate 1,3-D exposure to the following handlers:

- applicator (shallow shank w/ tarp)
- applicator (drip w/ tarp)
- applicator (drip w/o tarp)
- applicator (hand-wand)
- tarp remover

■ The exposure estimates for tarp removers were generated under the assumption that the handler is not wearing respiratory protection. Certain active product labels for CA omit requirements for respiratory protection for this scenario under certain conditions. Hence, the 10x factor for respiratory protection was left out, leading to higher exposure estimates.

■ There's increased uncertainty in the estimated breathing-zone air concentrations for handlers injecting Tri-Cal Trilone II. The label for this product lacks language, present on other applicable product labels, prohibiting the substitution of an enclosed tractor cab, equipped with filtration, for a respirator (a memo was sent to the registration branch to address this issue). A protection factor for a half-face respirator was incorporated into the exposure estimate calculations, reducing estimated breathing-zone 1,3-D concentrations. However, an enclosed tractor cab may not provide as much protection as the respirator, leading to breathing-zone 1,3-D air concentrations higher than those anticipated.

■ Use of the reentry worker exposure study data conducted by the registrant may have led to estimates higher than the actual exposure for this worker. The workers in the study reentered the treated field well before the end of the current CA 7-day REI: the exposure data for

the worker reentering the field after about 3.8 days, the longest interval of the study, was used to estimate exposure for this scenario.

■ The treated field acreages utilized to simulate the breathing-zone 1,3-D air concentrations for the occupational and residential bystanders may differ from that which is actually treated. This discrepancy could lead to under- or overestimation of actual exposures.

■ Both MCABLE and HEE5CB models based their exposure and, therefore, cancer risk estimates on predicted 1,3-D concentrations by SOFEA-2. However, when conducting the oncogenic risk estimations, HEE5CB employed a more stringent criterion for selecting the 1,3-D air concentrations and more restrictive assumption for evaluating the effect of residency-mobility on the exposed population than MCBALE. As a result, HEE5CB produced a higher (a factor of ~2) cancer risk estimates than MACBLE. Nevertheless, the exposure concentrations and residential-mobility assumptions employed by these two models are consistent with the real-life exposure conditions of 1,3-D. Hence, these models can provide a valuable insight into the range of exposures and oncogenic risks associated with the use of 1,3-D in California.

II. INTRODUCTION

A. CHEMICAL IDENTIFICATION

1,3-Dichloropropene (1,3-D) is a fumigant used to control nematodes, insects and disease organisms in soil. It promotes crop growth by minimizing competition with soil pests. 1,3-D has major uses in California in fruit and nut trees, strawberries, grapes, carrots and a host of other food and non-food crops (DPR Pesticide Use Report). It is commonly injected into soil on a preplant basis, after which it volatilizes and redissolves in the aqueous films that surround soil particles (Stott and Gollapudi, 2010). It is also applied through post-plant drip irrigation. Regardless of the application method, volatilization creates the opportunity for off-site transport and subsequent human exposure.

1,3-D formulations consist primarily of *cis* (E) and *trans* (Z) isomers in approximately equal proportions, as well as lower concentrations of other dichloropropenes and stabilizers. Some formulations also contain chloropicrin, dibromoethane or methylisothiocyanate, though neither of the latter compounds are found in formulations currently registered in California.

While the mechanism of pesticidal action is unclear, 1,3-D may work by inactivating vital enzymes through sulfhydryl or hydroxyl binding (Cox, 1992), or more generally, through formation of covalent bonds due to nucleophilic displacement. In mammals, including humans, it is an irritant, asphyxiant and sensitizer, though again, precise mechanisms of action are not identified (Liu, 2014).

The following human health assessment concentrates exclusively on risks emanating from inhalation exposure projected to occur in California under occupational, bystander and ambient scenarios. While the great majority of studies relevant to this evaluation were done by the Registrant to satisfy federal registration requirements, we also conducted a search for open-literature publications relevant to the characterization of mammalian inhalation toxicity using PubMed (http://www.ncbi.nlm.nig.gov/pubmed/) and the search terms “telone”, “telone II” and “1,3-dichloropropene”. Following an initial screen to remove duplicates, the combined searches identified 91 potentially relevant studies published between 1976 and 2011. Two additional publications were identified following a search of the NTP database (http://ntp.niehs.nig.gov/). Screening of these studies did not reveal data that added significantly to the Registrant data already on file at DPR.

B. REGULATORY HISTORY AND CURRENT REGULATORY LIMITS

1. U.S. EPA

In 1986, the U.S. EPA issued a Special Review of 1,3-D based on cancer concerns for workers. This review involved a Data Call-In (DCI) requiring additional residue chemistry, inhalation exposure, and environmental fate data. In December 1998, the U.S. EPA published a Reregistration Eligibility Decision (RED) for 1,3-D (USEPA, 1998). Following publication of

this RED, additional mitigations through label modifications were put in place including: lowered maximum application rates, deletion of some sites, closed loading requirements, technology to minimize spillage during application, improved stewardship materials, additional personal protective equipment (PPE), restricted entry interval (REI) increase to 5 days, soil moisture and sealing requirements, modified application methods, 300-foot buffer from occupied structures, loading requirements, ground water advisory, prohibition of use in northern states (MA, ME, MN, MT, ND, NH, NY, SD, UT, VT, WI) with shallow groundwater and vulnerable soils, 100-foot buffer between drinking water wells and treated fields, and prohibition of use in areas overlaying karst geology (USEPA, 2008b). Additional changes have been made including: changing the karst prohibition to prohibiting application within 100 feet of karst topological features, changing the buffer zone to 100 feet from occupied structures for some products, and establishing tolerance for grapes of 0.018 ppm when 1,3-D is applied by drip irrigation in established vineyards.

The U.S. EPA updated the human health assessment for 1,3-D in 2007 (USEPA, 2007). In addition, five other soil fumigants (methyl bromide, chloropicrin, metam sodium/potassium including methyl isothiocyanate (MITC), and dazomet were assessed in 2008. Although no new risks of concern were identified for 1,3-D, new safety measures were required for the other fumigants to increase protection for workers and bystanders. Label changes were required for 1,3-D products that contain chloropicrin. This affected 11 products registered for sale in California (Telone C-15, Telone C-17, Telone C-35, InLine, Pic-Clor 15, Pic-Clor 30, Pic-Clor 60, Pic-Clor 60EC, Tri-Form 30, Tri-Form 35, and Tri-Form 40/60). No changes were required for products containing only 1,3-D (Telone EC, Telone II, Telone II CA, and Tri-Cal Trilone II).

2. California

1,3-D is listed under Proposition 65 as a chemical known by the State of California to cause cancer. Due to this issue, between 1990 and 1995, California suspended all use of 1,3-D because unacceptably high levels were detected in a Merced County air monitoring program that was implemented under Assembly Bill 1807 (also known as the Toxic Air Contaminant Act of 1983). 1,3-D was reintroduced to the California market in 1995, and the Department of Pesticide Regulation imposed a limit on the total amount of 1,3-D that may be applied each year within a township. A township is a 6x6 mile area. The purpose of the township cap was to manage cancer risk associated with long-term inhalation exposure to 1,3-D. The township cap was set at 90,250 adjusted total pounds (ATP) of active ingredient per township-year. The term “adjusted” refers to adjustment factors used to multiply actual pounds of 1,3-D applied to account for flux differences dependent on fumigation method, month, and region. Application factors vary from 0.3x to 2.3x, and indicate the relative amount of 1,3-D that is potentially present in the air near treated fields. The higher the application factor value, the greater the proportion of the applied 1,3-D that may escape into the air (Verder-Carlos, 2014).

Following suspension of 1,3-D use in 1990, DowElanco (later known as Dow AgroSciences, DAS) conducted field trials under a variety of scenarios to monitor airborne concentrations of

1,3-D (Houtman, 1992; Houtman, 1993; Ray and Houtman, 1995). The results from these field trials have been used over the years to model air dispersion patterns associated with specific application methods and mitigation measures. In 1992 and again in 1996, 1,3-D labels were modified to minimize exposure to workers and residents living near treated fields and to include Worker Protection Standard (WPS) requirements.

Between January 1999 and the present time, there have been numerous permit conditions in California which affect the use of 1,3-D within the state. In June 2001, Enforcement Letter (ENF 01-31) set a minimum buffer zone of 100 feet from the application perimeter for any occupied residences, schools, hospitals and other similar sites (CDPR, 2001). It set a restricted entry interval (REI) for all application methods at 7 days after the application. It set conditions for application of 1,3-D when applied by either mechanical soil injection or drip application systems. In August 2001, ENF 01-31 was superseded by ENF 01-40 which defined a new set of permit conditions for 1,3-D use (CDPR, 2001). The changes in ENF 01-40 compared with ENF 01-31 included: a pending application factor for areas outside the San Joaquin Air Basin in December or January when the application depth is equal to or greater than 18 inches, elimination of the lower maximum application rate when a tarpaulin is used, and including maximum application rates for mechanical soil injection of both 94% and 97.5% Telone II products. In September 2002, ENF 01-40 was superseded by ENF 2002-037 (CDPR, 2002). New information included: clarification of use of buffer zones, provision of a finalized application factor when applying 1,3-D outside the San Joaquin Air Basin, and reference to the guidelines of the California Management Plan: 1,3-Dichloropropene (CDPR, 2002a).

3. The California Management Plan

In the "California Management Plan: 1,3-Dichloropropene", the California Department of Pesticide Regulation (CDPR) allowed Dow AgroSciences to restructure its seven year program to manage use of 1,3-D (CDPR, 2002a). This plan defined how the annual township caps (90,250 adjusted pounds 1,3-D per year per township) would be used. It allowed townships to "bank" unused annual allocation allowances since 1995. It also allowed townships to exceed the annual allocation limit and use up to 180,500 pounds annually as long as the 90,250 pound annual average was not exceeded. Use was to be validated by the California Data Management Systems (CDMS).

4. Current Permit Conditions

Current permit conditions can be found in the Department of Pesticide Regulation document: "Volume 3, Restricted Materials and Permitting, Pesticide Use Enforcement Program Standards Compendium" at: (http://www.cdpr.ca.gov/docs/enforce/compend/vol_3/rstrct_mat.htm) (CDPR, 2015c). The California permit conditions for 1,3-D set the maximum allowable application rate at 332 pounds of 1,3-D active ingredient per acre. The California permit conditions for chloropicrin with 1,3-D also limit the application block size to 40 acres at one location within a 24-hour limit period and set the minimum buffer zone at 100 ft for non-totally impervious film (TIF) tarp and untarped applications [Appendix K: Chloropicrin and Chloropicrin in

Combination with Other Products (Field Fumigant) Interim Recommended Permit Conditions]. These restrictions do not apply to 1,3-D products that do not contain chloropicrin (Telone II, Telone EC and Tri-Cal Trilone II).

5. Risk Management Directives and Related Correspondence

There are three documents and a cover letter for a memorandum from DPR scientists which provide Risk Management guidance regarding the use of 1,3-dichloropropene. A letter issued by CDPR deals with interim changes to buffer zones for 1,3-D applications to tree and vine crops (Gosselin, 2001a). A review by DPR scientists of the Dow proposal to shorten the buffer zone duration from 7 to 4 days for tree and vine applications explained why DPR retained the 7-day buffer zone duration (Johnson, 2001). On April 9, 2001 DPR issued a Risk Management Directive on "Managing 1,3-Dichloropropene (Telone) Chronic Risks" (Gosselin, 2001b). This directive set the acceptable oncogenic lifetime (70 year) risk at $1x10^{-5}$ at the 95^{th} percentile for 1,3-D. It also stated that the township cap would be maintained at the current level set by the permit conditions in effect in 2001. In 2002, DPR set forth actions needed for the future regulation of 1,3-D (Gosselin, 2002). This memorandum laid out the need for updating the Risk Characterization Document on 1,3-dichloropropene (CDPR, 1997a).

DPR suspended the approval of township cap exemptions until further notice in February of 2014 (Leahy, 2014).

DPR completed risk characterization documents (RCDs) in 1994 and 1997 that evaluated the human health risks associated with 1,3-D's projected application scenarios in California (CDPR, 1994; CDPR, 1997a). Critical regulatory endpoint values for acute, subchronic and chronic toxicity, as well as for oncogenicity were established in their documents. In 1997, DPR revised the acute endpoint in a regulatory memorandum (CDPR, 1999). All of DPR's regulatory endpoints determined prior to the current evaluation are listed below in Table II.1. Parallel regulatory endpoint values from USEPA are summarized in section V.D. below.

Table II. 1 1,3-D inhalation regulatory endpoint values from CDPR (1997)[a]

Exposure scenario Animal	NOEL (except as indicated)	LOEL	Toxic sign(s)	Reference
Acute **Fischer 344 rat**	ENEL = 77.5 ppm (≈54.8 mg/kg) [b]	775 ppm	Salivation, lacrimation, lethargy, urine/fecal staining	(Streeter *et al.*, 1987)
Acute (CDPR, 1999) **CD rat**	10 ppm [c]	60 ppm	Body weight decrement	(Gollapudi *et al.*, 1998)
Subchronic **Fischer 344 rat**	10 ppm (≈7.1 mg/kg/day) [d]	30 ppm	Nasal epithelial histopathology	(Stott *et al.*, 1984)
Chronic **B6C3F1 mouse**	5 ppm [c]	20 ppm	Nasal epithelial and bladder histopathology	(Stott *et al.*, 1987)
Oncogenicity **B6C3F1 mouse**	Q_1 (MLE) [e] = 0.0035 (mg/kg/day)$^{-1}$ Q_1* (UCL) [e] = 0.0079 (mg/kg/day)$^{-1}$		Bronchioloalveolar adenomas	(Stott *et al.*, 1987)

[a] The only exception is the revised acute endpoint established in CDPR (1999) (3rd row of table).

[b] Conversion to an equivalent internal dose was based on the following calculation:

Internal dose = (LOEL in ppm/10) x purity x (4.53 mg/m^3) x (rat breathing rate) x (4 hr /24 hr)

77.5 x 0.975 x 4.53 x 0.96 m^3/kg/day x 4/24 = 54.8 mg/kg

[c] Conversion to an equivalent internal dose was not carried out for these scenarios.

[d] Conversion to an equivalent internal dose was based on the following calculation:

Internal dose = (NOEL in ppm) x purity x (4.53 mg/m^3) x (rat breathing rate) x (6 hr /24 hr) x (5 d / 7 d)

10 x 0.909 x 4.53 x 0.96 m^3/kg/day x 6/24 . 5/7 = 7.1 mg/kg/day

[e] MLE: maximum likelihood estimate of oncogenic potency; UCL: upper confidence limit

6. Exposure limits generated by other regulatory bodies

The American College of Governmental Industrial Hygienists listed 1 ppm as its 8-hr TLV for 1,3-D. The National Institute of Occupational Safety and Health also listed 1 ppm as its 10-hr TLV (ATSDR, 2008). An inhalation reference concentration (RfC) of 0.02 mg/m^3 (~0.0045 ppm) was established by USEPA based on benchmark concentration analysis of the incidence of hypertrophy and hyperplasia of the respiratory epithelium in a mouse chronic study. ATSDR set minimal risk levels (MRLs) of 0.008 and 0.007 ppm for intermediate and chronic exposure, also based on hypertrophy and hyperplasia of the respiratory epithelium. They did not establish an acute MRL. The European Food Safety Authority set an "inhalatory acceptable operator exposure concentration (AOEC)" of 0.1 ppm based on the 10-ppm NOAEL (hyperplasia of the nasal respiratory epithelium at 30 ppm) from the 13-week rat inhalation toxicity study (EFSA, 2009).

C. PRODUCT FORMULATIONS AND USES

As of August 2015, there were 17 registered products containing 1,3-D in California, including one product intended solely for manufacturing or reformulation use. 1,3-D-containing products are available in both pressurized and non-pressurized containers, as compressed liquids in cylinders or liquid solutions containing emulsifiers. Many are mixtures with chloropicrin. 1,3-D products are used for pre-plant soil fumigation for many crops, using injection equipment or drip irrigation. Table II.2 summarizes the 1,3-D injection and drip irrigation products registered in California, and the respective active ingredient contents.

Table II. 2 1,3-D-Containing Products as of August 2015

Application	Product	% 1,3-D	% chlor[a]
Injection	Pic-Clor 15	82.9	14.9
	Pic-Clor 30	68.3	29.8
	Pic-Clor 60	39.0	59.6
	Telone C-15	82.9	14.9
	Telone C-17	81.2	16.5
	Telone C-35	63.4	34.7
	Telone II	97.5	
	Tri-Cal Trilone II	97.5	
	Tri-Form 30	68.3	29.8
	Tri-Form 35	63.4	34.8
	Tri-Form 40/60	39.0	59.6
	Tri-Form 80	19.5	79.8
Drip	Telone EC	93.6	
	InLine	60.8	33.3
	Pic-Clor 60 EC	37.1	56.6
	Tri-Form 80 EC	18.5	79.8

[a] mixture containing chloropicrin

Pre-plant soil fumigation is done for many crops, using injection equipment (pressurized liquid) or drip irrigation (emulsified liquids). DPR describes three main types of pre-plant soil fumigation: broadcast fumigation (where the application of a pesticide occurs uniformly over the area to be treated without regard to arrangement of crops as in rows); strip fumigation (applications that have alternating fumigated and unfumigated areas, often with prior or subsequent fumigation of the unfumigated areas); and bed fumigation (where pre-formed beds are fumigated and the furrows are not) (CDPR, 2014). Additionally, a few soil injection products (Telone C-17, Telone C-35, Telone II, Telone II CA, and Tri-Cal Trilone II) can be used for tree planting sites using hand-held equipment.

1. Pesticide Usage

Two databases are available for investigating use of 1,3-D. The first is the Pesticide Use Report (PUR) database maintained by DPR while the other is the AGRIAN® PUR database generated by Dow Agrosciences. California requires reporting of all agricultural applications of pesticides, as well as other uses when pesticides are applied by a licensed applicator. These data are collected in DPR's Pesticide Use Report (PUR) database. The AGRIAN® PUR records are specific to 1,3-D and are part of the California Management Plan which was put in place to ensure that the amounts of 1,3-D applied in CA do not exceed the use limits set by DPR (CDPR, 2002a). The pesticide use records in the AGRIAN® database contain much more information specific to 1,3-D (e.g., specific method of application, application date, application company, application rate), than that provided in DPR's PUR database and are more up to date. Moreover, the records for the total pounds applied statewide for the latest 4 years (i.e., 2010-2013) of data on the DPR PUR database are in within 2.1 to 6.4% of the totals for these years on the AGRIAN® PUR database (Table II.3).

Table II. 3 Pounds of 1,3-D applied: comparison of annual statewide totals between the DPR and AGRIAN PUR databases

Year	DPR PUR database	AGRIAN® PUR database	% Difference
2010	8771323	8953350	2.1
2011	10907012	11197043	2.7
2012	12012976	11248926	6.4
2013	12917296	13216014	2.3
2014	no data	13775265	n/a

As a result, the bulk of the use seasons and the estimated seasonal application rates used for estimating exposure in this document were derived from the latest 5 years (2010-14) of use records in the AGRIAN® PUR database (DAS, 2011; DAS, 2012; DAS, 2013; DAS, 2014; DAS, 2015a).

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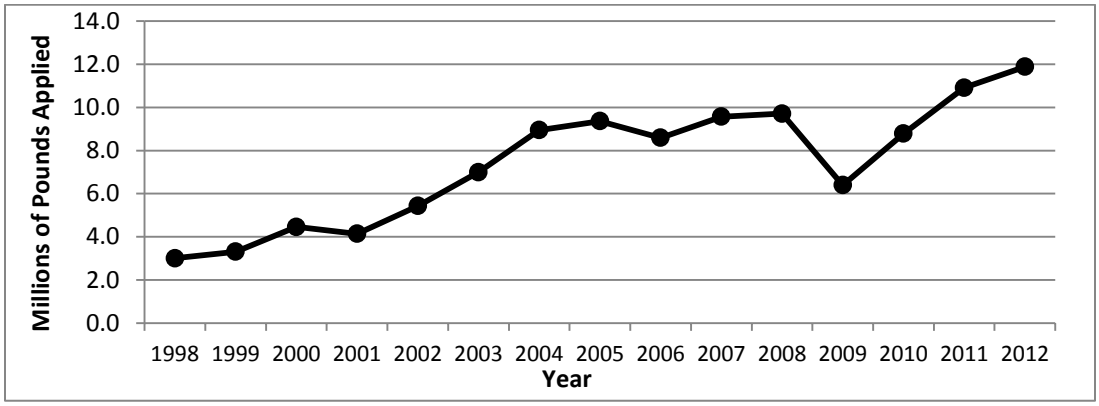
General Use Information

A query of the DPR PUR database for 1,3-D use in California from 1998 to 2012 shows a substantial increase in the use of 1,3-dichloropropene due, in part, to increased incidence of

resistance-breaking nematodes in nematode-resistant tomato cultivars and restrictions on the use of methyl bromide as a fumigant (CDPR, 2015d; CDPR, 2015e). In 2000, the total use of 1,3-dichloropropene in California was approximately 4.5 million pounds, but by 2011 use more than doubled to reach 10.9 million pounds for that year. Between 2004 and 2010, the pounds of 1,3-D used per year stabilized and varied between 8.6 and 9.7 million pounds per year. The one exception to this range was 2009, when use dropped to 6.2 million pounds per year, possibly in part to limited availability of 1,3-D. Variation in weather patterns and crops planted in 2009 may also have contributed to this low level of use which returned to 8.8 million pounds in 2010. As of 2012, use of 1,3-D had reached about 12 million pounds annually. Use of 1,3-D in terms of pounds applied annually from 1998 to 2012 is summarized in Figure II.1.

Figure II. 1 Summary of annual application of 1,3-dichloropropene from 1998 to 2012



For preplant soil fumigation, 1,3-dichloropropene is formulated as a concentrated liquid for drip irrigation or as a pressurized liquid for soil injection. According to the 1,3-D PUR report, roughly 80% of the 1,3-D applied over a recent 5 year period was soil injection products. The remaining portion was applied using drip irrigation products (InLine, Pic-Clor 60 EC, and Telone EC). This data is summarized in Table II.4.

Table II. 4 Percent average of 1,3-D applied using drip or soil injection applications

Application method	Pounds Applied by Year					5-year average (%)
	2008	2009	2010	2011	2012	
drip	1979592	1122792	2069845	2082575	2064571	19.6%
soil injection	7726970	5276723	6707247	8827592	9810106	80.4%

According to the AGRIAN PUR database, the bulk of the 1,3-D applied from 2010-14 was via deep shank injection. Well below the deep shank injection method are the drip and shallow shank methods. The other category shown in the AGRIAN database is “Unknown/Other”. This

category of application method accounted for the least number of pounds of 1,3-D applied during the 5-year period (Table II.5).

Table II.5 Percent of 1,3-D Applied Using Application Methods Listed in AGRIAN PUR database (2010-14) [a].

Application Method	Pounds of 1,3-D Applied	% of Total
Deep Shank	45,413,479	78
Drip	7,380,226	13
Shallow Shank	4,871,175	8
Unknown/Other	725,718	1

[a] For the deep shank, drip, and shallow shank application methods, the numbers of pounds applied and percentages are for applications done with and without the use of a tarp.

In California, 1,3-D is registered for agricultural use only as a preplant fumigant for many food and feed crops including: fruit and nut crops, vegetable crops, field crops, and nursery crops. According the PUR report for 1,3-D-use from 2008 to 2012, 20.5% of total 1,3-D used did not specify the crop to be planted after fumigation (CDPR, 2015e). For the remaining 79.5% of 1,3-D use, the crop was specified and the following crops account for most of the 1,3-D used: fruit and nut crops, strawberries, grapes (all types), carrots, sweet potatoes, tomatoes (all types), peppers, melons (all types), potatoes, Brussels sprouts, raspberries, and leafy greens. In addition, "other" crops together account for the remaining 1.4% of total use and separately account for less than 0.5% of the annual use. Other crops include onions; corn, wheat, oats, and alfalfa fodder; blueberries; beans; beets; bitter melon; broccoli; cauliflower; eggplant; asparagus; leeks; squash; and taro. Table II.6 summarizes the use of 1,3-D by commodity between 2008 and 2012. Use in greenhouses and other enclosed areas is explicitly forbidden by California permit conditions (CDPR, 2015c).

Table II. 6 Use of 1,3-Dichloropropene by crop for 2008-2012

Crop	Pounds applied by year[a]					5-year Total	Percent of Total
	2008	2009	2010	2011	2012		
Fruit and Nut Trees[b]	2202576	1817078	2404478	2958937	1838825	11221894	23.5%
Strawberries	1793309	1135408	1952932	2265219	2773871	9920738	20.8%
Crop Not Specified[c]	1759657	1209564	1387134	2180465	3228694	9765513	20.5%
Grapes, all types[d]	831818	631984	820581	1576214	1107433	4968030	10.4%
Carrots	1032096	392443	603962	400810	909092	3338404	7.0%
Sweet Potatoes	609251	428880	646359	480551	730529	2895570	6.1%
Tomatoes, all types[e]	591768	306059	372281	198789	240732	1709630	3.6%
Peppers	256938	84261	102651	133777	119833	697460	1.5%
Melons, all types[f]	177820	96294	164243	102508	101533	642398	1.3%
Potatoes	113634	176755	112848	98819	108856	610912	1.3%
Brussels Sprouts	70516	11957	48656	172289	195528	498946	1.0%
Raspberries	13955	13166	29444	92908	271904	421376	0.9%
Leafy Greens[g]	68278	15968	56148	50504	104844	295742	0.6%
Other[h]	184945	79698	75376	198378	143003	681402	1.4%

[a]CDPR (2015e) Crops are arranged in descending order indexed by the 5 year total

[b]**Fruit and Nut Trees** includes: almond, apple, apricot, cherry, citrus, fig, grapefruit, lemon, nectarine, olive, orange, peach, pear, pecan, persimmon, pistachio, plum, pomegranate, prune, stone fruits, tangelo, tangerine, and walnut.

[c]**Crop Not Specified** includes: soil, uncultivated agricultural areas, N_OUTDR transplants, N_OUTDR plants in containers, N_GRNHS plants in containers, N_OUTDR flowers or greens, blank, and N_GRNHS flowers or greens

[d]**Grapes** includes: wine grapes and other grapes

[e]**Tomatoes** include: processing tomatoes and other tomatoes

[f]**Melons** include: cantaloupe, melon, and watermelon

[g]**Leafy Greens** include: bok choy, cabbage, Chinese cabbage, head and leaf lettuce, kale, parsley, and spinach

[h]**Other** are crops which represent 0.5% or less of the yearly application. These include: onions; corn, wheat, oats, and alfalfa fodder; blueberries; beans; beets; bitter melon; broccoli; cauliflower; eggplant; asparagus; leeks; squash; and taro

The 2008-2012 PUR reports were also used to obtain monthly application amounts of 1,3-D to characterize seasonal use. Seasonal use is defined by DPR as use which is greater than 1 week but significantly less than one year. Specifically, the length of the season was calculated by summing the number of months having application amounts equal to or greater than 5% of the annual total. To be defined as a use season this sum must be at least 3 months. The top five counties in which 1,3-D was used in the 5-year interval 2008 – 2012 are Fresno, Kern, Monterey, Merced, and Tulare; together, they accounted for over 50% of statewide use (CDPR, 2015e). Table II.7 shows the pounds applied for each of the 5 top counties for 2008-2012, and respective percent usage of total pounds applied in California.

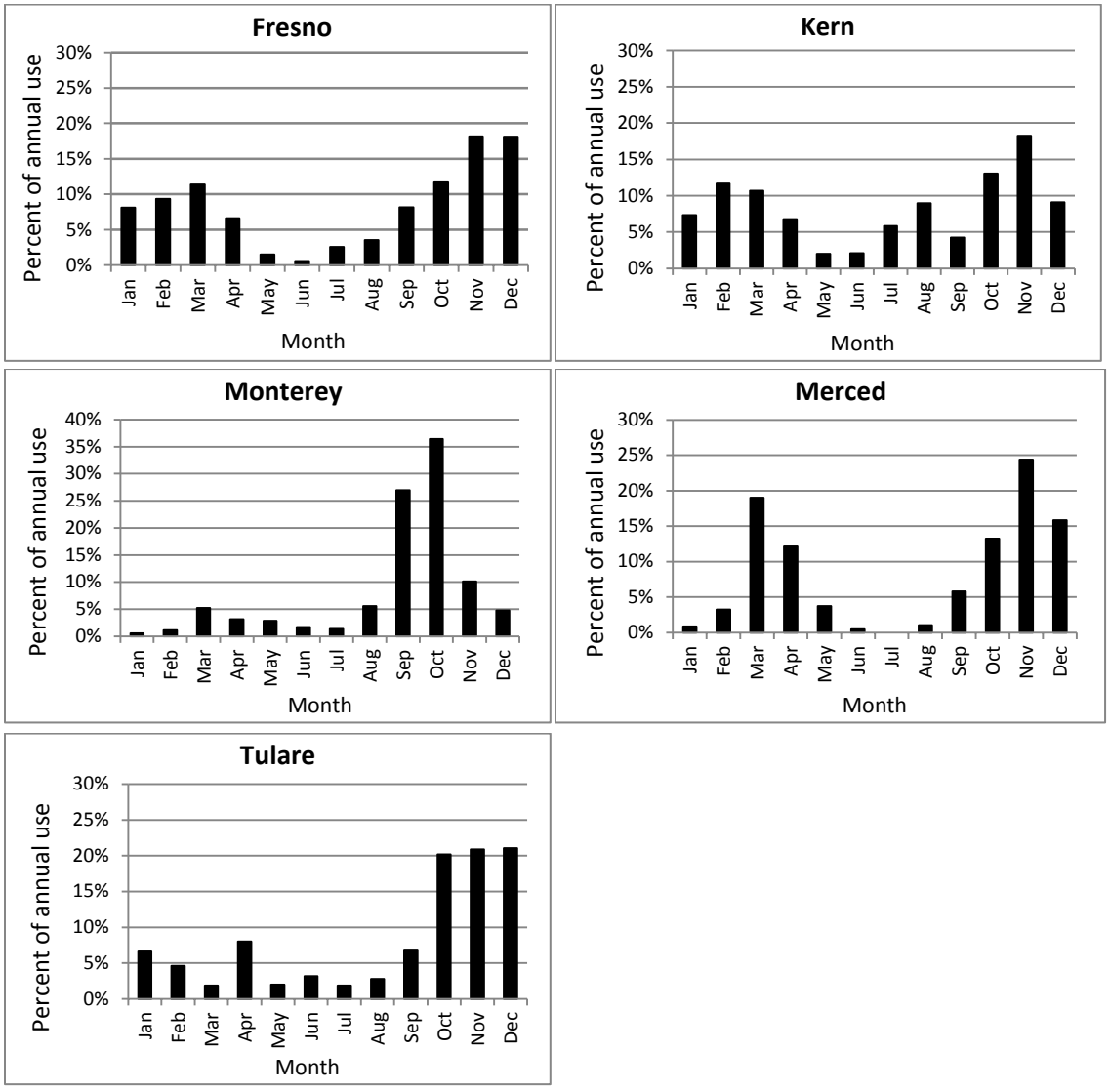
Table II. 7 Total Pounds of 1,3-D Applied in the Top 5 Counties

County	Pounds Applied (2008-2012)	Percent of Total[a]
Fresno	6617008.2	13.9%
Kern	6258636.5	13.1%
Monterey	4627262.9	9.7%
Merced	4613866.5	9.7%
Tulare	3799171.6	8.0%

[a] Total 1,3-D applied in California 2008-2012: 47,668,014.4lbs

Figure II.2 summarizes the monthly percentage of total 1,3-D used annually over a recent 5 year period in each of these 5 counties. The seasonal patterns within the top 5 counties ranged from 4 to 9 months, with an average of 8 months.

Figure II. 2 Seasonal application patterns of 1,3-D: average monthly use by top 5 highest-use counties



Use Seasons and Estimated Seasonal Application Rates Used for Estimating Exposure

Use seasons and estimated seasonal application rates were generated, using AGRIAN® PUR data from 2010-14, for the applicator, loader, tarp remover, reentry worker, and residential bystander exposure scenarios. For the reentry worker and residential bystander, the use season and seasonal application rate were based upon the highest use county for each application method. Determining the actual number of application days within the use season allowed for greater resolution of seasonal exposure (i.e., SAC). Determining the number of application days within the year allowed for greater resolution of annual exposure (i.e., AAC). The seasonal application rate for each method consists of the median of the application rates in the database from 2010-14. The estimated season application rates used for the reentry worker were also applied for estimating residential bystander exposure. Use seasons and estimated seasonal application rates were also estimated for the handler scenarios, the detail of the use data allowed for estimation of the use season and seasonal application rate in the highest use county for the company applying the greatest amount of 1,3-D using shallow shank, deep shank, or drip. Determining the actual number of application days within the use season allowed for greater resolution of seasonal exposure (i.e., SAC). Determining the number of application days within the year allowed for greater resolution of annual exposure (i.e., AAC). The seasonal application rate for each method consists of the median of the application rates in the database from 2010-14 (Table II.8.).

Table II. 8 Use seasons and seasonal application rates derived from

AGRIAN® PUR data base (2010-14)

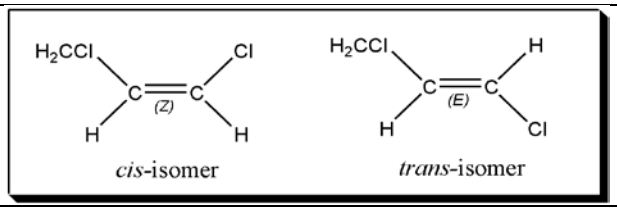
Method	Use season (months)	Application days in use season	Application days in year	Seasonal application rate (lbs/acre)
Handler Exposure Scenarios				
shallow shank	3 mons (Aug-Oct)	85	115	154
deep shank	8 mons (Jan-Mar, Aug-Dec)	193	228	327
drip	3 mons (Aug-Oct)	73	121	101
Reentry Worker and Residential Bystander Exposure Scenarios				
shallow shank	4 mons (Aug-Nov)	108	159	153
deep shank	7 mons (Jan-Mar, Sep-Dec)	203	283	327
drip	3 mons (Aug-Oct)	82	163	103

The use data for applications conducted with and without the use of tarps were combined. Although, the last two years of the PUR database (i.e., 2013-14), clearly show which applications utilized tarps and which did not, the previous 3 years of records do not. Hence, the data for both types of applications were combined for estimating use seasons and seasonal application rates.

D. CHEMICAL DESCRIPTION AND PHYSICO-CHEMICAL PROPERTIES

1,3-D is a chlorinated hydrocarbon. Formulations consist of approximately equal parts of the "Z" (*cis*) and "C" (*trans*) isomers. Physical and chemical properties of this white or amber liquid appear in Table II.9 below.

Table II. 9 Physical and chemical properties of 1,3-dichloropropene from Stott and Gollapudi (2010) and CDPR (2012)

Property	Value
Chemical name	1,3-dichloropropene
Common names	Telone II, 3-chloropropenyl chloride, alpha-chloroallyl chloride
CAS registry number	1,3-D: 542-75-6 *Cis*-isomer: 010061-01-5 *Trans*-isomer: 010061-02-6
Physical state; color	Liquid; white or amber
Odor	Sweet, penetrating, chloroform-like
Molecular formula	$C_3H_4Cl_2$
Molecular structure	
Molecular weight	110.98 g/mole
Chemical family	Chlorinated hydrocarbon
Boiling point	*Cis*-isomer: 104ºC *Trans*-isomer: 112ºC
Flash point	28ºC
Melting point	-84°C
Vapor pressure	28.0 mm Hg (20°C) 34.3 mm Hg (25°C)
Water solubility	~2 g/kg
Henry's Law constant	3.55 x 10^{-3} atm·m^3/mol
Log K_{ow}	1.82 (miscible with acetone, benzene, carbon tetrachloride, heptane & methanol
Log K_{oc}	*Cis*-isomer: 1.36 *Trans*-isomer: 1.41
Density	*Cis*-isomer: 1.22 g·cm^{-3} *Trans*-isomer: 1.22 g·cm^{-3}
Conversion Factor	1 ppm = 4.53 mg·m^{-3} at 25^{o}C

E. ENVIRONMENTAL FATE

The environmental fate of 1,3-D was examined in detail in a previous CDPR report authored by Edgar Vidrio (CDPR, 2012). The following summary is distilled from that document.

1. Soil

The half-life ($t_{1/2}$) of 1,3-D in soil varies with soil type, temperature and microbial content, but can range as high as 69 days. Interestingly, the soil $t_{1/2}$ is lower under anaerobic conditions, ranging between 2.4 and 9.1 days. Adsorption of 1,3-D to soil is stronger in the vapor phase, which dominates at normal soil temperatures, than in water. Diffusion (as opposed to leaching) is the primary mode of soil mobility. Hydrolysis in soil is influenced by moisture content and organic matter, and follows pseudo first-order kinetics to yield chloroallyl alcohol and other degradates. While runoff rates are low due to high soil persistence and high hydrolysis rates, 25-56% of applied 1,3-D will volatilize within 2 weeks.

2. Water

The volatilization $t_{1/2}$ of 1,3-D from surface water was determined to be ~4 hours based on modeling of a river, though its maximum estimated $t_{1/2}$ is 50 hours when the model is of a pond and includes adsorption to particulates . Hydrolysis in water occurs over periods of days to weeks. Photolysis occurs over a much longer period, so is not a significant factor in 1,3-D degradation. 1,3-D is unlikely to be a groundwater contaminant.

3. Air

As noted, 1,3-D readily volatilizes into air---about 40% within 2-3 weeks---thus creating the potential for inhalation exposure. The tropospheric $t_{1/2}$ of the *trans* and *cis* isomers is 30 and 50 hours, respectively. Decomposition is aided by atmospheric particulates, light intensity and NO_2 concentration. In addition, airborne 1,3-D is degraded by hydroxyl radicals ($t_{1/2}$ = 7 hours and 12 hours for the *trans* and *cis* isomers, respectively) and by ozone ($t_{1/2}$ = 12-52 days). 1,3-D is also photooxidized to 3-chloropropionyl chloride and subsequently hydrolyzed to 3-chloropropionic acid, which is removed by rainfall.

4. Vegetation

1,3-D applied to soils usually dissipates before planting, which, in addition to its rapid metabolism in cases when it *is* absorbed, makes it unlikely that residues will be found in food crops. The measured Log K_{OW} of 1.82 does not indicate a potential for bioaccumulation in aquatic and terrestrial food chains.

III. TOXICOLOGY PROFILE

A. PHARMACOKINETICS

1. Inhalation, human

A human study demonstrated efficient uptake of 1,3-D vapor, a transitory presence of parent 1,3-D in the blood, and substantial urinary excretion of products of glutathione-derived products in the urine ((Waechter *et al.*, 1992)). Six male volunteers were exposed by inhalation for 6 hours to 1 ppm of *cis*/*trans*-1,3-dichloropropene (1,3-D, Telone II Soil Fumigant), consisting of 50.6% *cis*-1,3-dichloropropene and 45.2% *trans*-1,3-dichloropropene. Investigators assessed concentrations of the above 2 isomers in exhaled air and blood during and after exposure, as well as urinary excretion of two key conjugation products: the unaltered *cis* and *trans*1,3-D: namely *cis* or *trans* N-acetyl cysteine's (1,3-D NAC's).

Respiratory uptake was about 80% for both isomers (based on comparison of chamber air with exhaled air). There was a rapid "plateau" of concentrations of both isomers in exhaled air in most subjects during exposure, there being no systematic change in exhaled air concentration from the first exposure period measurement from the first 5 minutes of treatment. There was a rapid decrease in exhaled air concentration after termination of exposure (reduction to about 5% of mean exposure period levels at 5 minutes after dosing), indicating that investigators found no perceptible off-gassing from the blood compartment after cessation of treatment. Subjects absorbed estimated total amounts of 4.2 mg of *trans* and 4.6 mg of *cis* isomers.

Blood concentrations achieved a near steady state within about 10 minutes, averaging about 0.7 ng/g for *cis* and about 1.3 ng/g for *trans* Telone, respectively, for most samplings during the exposure period. These levels were about twice the respective limits of detection for respective isomers, limiting the resolution of this study. Blood concentrations of the 1,3-D isomers varied appreciably between subjects, although inter-subject differences were minor for 4 of the 6 subjects for the first 3 treatment hours. One subject had mostly levels below quantification limits during the exposure phase. Several subjects had appreciably higher blood concentrations of *cis* and/or *trans* 1,3-D at 6 hours compared to earlier samplings. One particular subject had maximal (6-hr) blood levels of both isomers that were markedly higher (up to about 3x) compared to the other subjects as well as compared to his own levels in earlier samplings. Post-exposure blood concentrations of *cis*- or *trans*-1,3-D in most subjects were generally below detection limits or were marginally quantifiable levels within 15 minutes, with occasional detectable levels in some subjects for at least 4 hrs for one or both isomers. The subject with the highest concentrations of both isomers at the final treatment phase assessment still had detectable levels of both isomers 4 hrs into the recovery period.

A substantial proportion of dose was metabolized by initial glutathione conjugation of parent 1,3-D, eventually presenting as isomer-specific N-acetylcysteines (1,3-D NAC's). Total urinary elimination by assayed 1,3-D NAC's was quite variable: *cis*-1,3-D NAC excretion ranged from 2.3 to 5.8 mg, whereas *trans* -1,3-D-NAC ranged from 0.4 to 1.9 mg. About 44% of the mass of

those conjugates was attributable to parent Telone, and only these were characterized and quantified in this study. Thus, although the 1,3-D NAC conjugates constituted large portions of absorbed Telone II, particularly for the *cis* isomer, fate of much of absorbed Telone II was not addressed in this study. Initial phase half-lives for urinary excretion of *cis* and *trans* 1,3-D NAC's were 4.2 and 3.2 hrs, respectively. Terminal phase half-lives were 12.3 and 17.1 hrs, respectively. Although the 1,3-D NAC's are suitable indicators of exposure at the relatively high levels tested, sensitivity of analysis at that time (1992) was insufficient to assess the much lower anticipated exposure levels of field workers and particularly of bystanders.

When investigators put together the information (per subject and per isomer) on (1) absorbed dose (based on exhaled air concentration vs. exposure concentration) and (2) excreted dose (based exclusively on 1,3-D NAC in urine), they concluded that about 75% of absorbed dose of the *cis* isomer was excreted as a direct conjugation product of the *cis* parent, whereas only about 23% of absorbed dose of the *trans* isomer was excreted as a direct conjugation product of the *trans* parent.

This study was considered supplementary (i.e. valid and useful, but not designed to address a FIFRA data requirement). Results showed the usefulness of NAC metabolites as exposure indicators, and demonstrated distinct differences between the fates of the two 1,3-D isomers. Invaluable in many ways, this study provided no information on Phase I metabolism in humans, which must be determined from animal studies.

2. Inhalation, rat

The primary rat inhalation-route disposition study confirmed that the rat behaves similarly to humans in terms of absorption and disposition of parent isomers in blood. In addition, this study determined that most absorption was in the lung rather than in the upper respiratory region, and assessed features of saturability of non-protein sulfhydryl conjugation capacity (such as glutathione) in key tissues. Anesthetized male F344 rats were exposed to 1,3-D, 92.1% purity (49.3% *cis* and 42.3% *trans*) by nose-only inhalation exposure at 30, 90, 300, or 900 ppm for 3 hours for most experiments ((Stott and Kastl, 1986)).

Based on breathing rate, tidal volume, and measurements of 1,3-D levels in air entering and leaving the head-only exposure space, estimated absorbed dose was 82% of inhaled 1,3-D for 30 ppm exposure, and 62-65% for 90-900 ppm.

For blood analysis of *cis* and *trans* 1,3-D, investigators repeatedly sampled blood from an indwelling jugular cannula. At 30-90 ppm, blood levels at 1, 2, and 3 hours of exposure were constant, followed by rapid declines at the end of exposure (especially rapid for *cis* isomer). At 300 ppm and particularly at 900 ppm, blood levels rose markedly from hour to hour during the exposure period, indicating a saturation of metabolism or excretion processes at very high dose levels. Estimated phase 1 elimination half-lives for *cis* 1,3-D were 3-5 min at 30 to 300 ppm, and 14 min for 900 ppm. Corresponding elimination half-lives for *trans* 1,3-D were about 5 min for

lower dose levels, and 27 min for 900 ppm. *Trans* 1,3-D, although not as abundant in the technical as the *cis* isomer, was consistently more abundant in blood during exposure, and was more slowly cleared after cessation of dosing. Blood plateau levels at 30 ppm were 0.085 and 0.12 µg/ml for *cis* and *trans* 1,3-D, respectively. The most profound difference in blood concentrations of isomers at termination of exposure was in 300 ppm rats, where the *trans* isomer was over twice the concentration of the *cis* isomer. Thus it is apparent that the *cis* isomer of 1,3-D is more readily cleared from the blood than the *trans* isomer, although this particular study did not identify the eventual metabolites.

Investigators determined the proportion of absorption in upper and lower respiratory tract (URT and LRT, respectively) after sectioning and catheterizing the tracheae of anesthetized rats. LRT absorption was assessed by analyzing inhaled and exhaled gasses in the isolated caudal portion of the endotracheal tube. URT uptake was estimated by examining input and output gas 1,3-D in a direct unilateral flow model. Sums of LRT and URT uptakes were compared against absorbed dose in similarly anesthetized rats with intact respiratory tracts. These tests evaluated 90 ppm and 150 ppm 1,3-D test atmospheres only. Investigators determined that 73-79% of total absorbed 1,3-D was absorbed in the LRT, the balance of absorbed 1,3-D in the cannulated rats being absorbed in the URT. The sums of these two isolated respiratory tract uptake estimates were acceptably close to measured absorption in the anesthetized intact rat. When expressed as percent of theoretical uptake, LRT and nose-only intact rats in this phase of the study absorbed about 50% of available 1,3-D. About 11-16% of dose was absorbed in the URT.

Respiratory frequency and tidal volume were assessed with a pressure transducer designed to measure pressure changes in the head-only space. There was a consistent decrease in respiratory rate (breaths/min) with increasing dose, with no consistent change in tidal volume, hence there was a decrease in respiratory minute volume at 300 to 900 ppm. A combination of reduced respiratory minute volume and reduced percent of dose absorbed at the highest dose resulted in a reduction of uptake/(exposure concentration) to less than 50% at 900 ppm compared to 30 ppm.

Investigators estimated tissue non-protein sulfhydryl (NPSH) content in homogenates of liver, kidney, or lung immediately after dosing with 0 or 90 ppm 1,3-D (tissue proteins were removed by precipitation with trichloroacetic acid or *m*-phosphoric acid). Liver and kidney NPSH contents were statistically significantly reduced by 1,3-D: 31% reduction of NPSH in kidney and 41% decrease in liver. There no reduction NPSH observed in lung. Given that most human exposures would be well under 90 ppm, there is little reason to expect significant reduction of NPSH content in tissues of exposed humans.

3. Oral, rat

A repeat-dose gavage study in rats confirmed presence of mercapturic metabolites as shown in other studies, but also found subsequent oxidation of the conjugates to sulfoxides and sulfones, and demonstrated that there was significant degradative metabolism of the parent molecule as evidenced by considerable CO_2 in exhaled air. Five rats/sex were dosed daily with 5 mg/kg 1,3-

dichloropropene (1,3-D), (54.2% *cis* and 45.8% *trans*), by gavage in corn oil (5 ml/kg) for 14 consecutive days ((Waechter and Kastl, 1988)). On day 15, rats were fasted for 8 hr prior to dosing with 5 mg/kg of *cis/trans*-1,3-dichloropropene, uniformly labeled with ^{14}C, which assayed prior to dosing at 96.3% 1,3-D (53.5% *cis* and 43.0% *trans*). Two additional fasted rats/sex, which had no prior 1,3-D treatment, were treated with the same labeled material. Urine, feces, exhaled CO_2, other expired volatiles, and tissue levels were assessed. Sacrifice was 48 hrs after labeled treatment.

Chromatographic resolution was not sufficient to routinely separate the *cis* and *trans* isomers of mercapturic acid conjugates, and also not sufficient to separate subsequent sulfoxide and sulfone products from one another. Other metabolites were not quantified; nonetheless, the authors proposed that 1,3-D which did not undergo direct conjugation of parent molecules was likely metabolized to 3-chloroallyl alcohol, then to its aldehyde and acid derivatives, and eventually to release of CO_2 and incorporation of the residues into the TCA cycle.

Approximately 62-65% of administered dose was found in urine, 26% in exhaled CO_2, 5% in feces, and 4-6% in tissues and carcass, with no observed sex difference. At 48 hrs after dosing of pre-treated rats, highest concentrations of label (about 1 µg equivalent/g tissue) were in bladder and forestomach. Lowest concentrations were in brain and fat (less than 0.1 µg equivalent/g tissue), with about 0.2 to 0.4 µg equivalent/g in other tissues. Urinary excretion of the *cis/trans* N-acetyl cysteine conjugates comprised 26-28% of administered dose. The combined close-eluting sulfoxide and sulfone residues comprised about 14% of the administered dose. Most excretion occurred within the first 12 hours. There were no clear differences in disposition between sexes or resulting from 14-day pre-treatment vs. naïve rats. This is a valid supplemental study.

4. *In vivo* and *in vitro* pharmacokinetics, mouse

A published report showed that epoxides were generated in measurable amounts *in vitro* and *in vivo* from 1,3-D, and that these epoxides and their common derivative, 3-chloro-2-hydroxypropanal, were potent mutagens (Schneider *et al.*, 1998a). The 92% cis/*trans* (1:1) 1,3-dichloropropene (1,3-D) obtained for this study contained 0.2% each of *cis* and *trans* epoxides, which was quantitatively removed by DMSO at room temperature (*i.e.*, degraded by this solvent without the need for added cofactors) in studies showing epoxide formation.

The metabolic oxidation of epoxide-free 1,3-D [*cis*/*trans* (1:1)] to form respective epoxides was demonstrated in Swiss-Webster mice (700 mg/kg by intraperitoneal injection). Liver extracts of these mice yielded several times more *cis*- epoxide than *trans*- epoxide. *Cis* and *trans*- epoxide levels in these mice livers were several hundred-fold lower than corresponding 1,3-D levels. *In vivo* treatment of mice with either purified *cis*- or *trans*-1,3-D yielded only the corresponding *cis*- or *trans*- epoxide *in vivo* (measured in extracts from mouse liver). This same specificity was also shown *in vitro* (in mouse liver microsomes). In livers of mice dosed *ip* with epoxide-free 1,3-D [*cis*/*trans* (1:1)], *trans*- 1,3-D levels predominated over *cis* (roughly 2-fold) throughout the

150 minute sampling period. In contrast, *cis*- 1,3-epoxide predominated over the *trans*-epoxide in the same liver extracts by about 2-fold during that period.

Similarly, *in vitro* mouse liver microsomal preparations of epoxide-free 1,3-D in the presence of NADPH also yielded preferentially *cis* epoxide. When GSH plus NADPH were added to the same *in vitro* system, there were markedly reduced *cis* and *trans* epoxide levels, and respective parent 1,3-D levels in liver extracts were reduced to about 30% of respective isomer levels compared to microsomal preparations lacking NADPH and GSH. Potential metabolites sought but not found in this assessment were 2-chloroacrolein and *cis*- and *trans*-3-chloroacrolein.

Cis/trans 1,3-D epoxide decomposition in pH 7.4 buffer yielded predominantly 3-chloro-2-hydroxypropanal (including its dimer). Whereas both epoxides degraded spontaneously in pH 7.4 buffer, addition of GSH roughly doubled degradation rate, and GSH plus glutathione S-transferase (GST) increased the latter rate about 60-fold.

Investigators noted that *cis/trans* 1,3-D epoxides dissolved in DMSO yielded a virtually quantitative product of 2-chloroacrolein within 280 minutes at 22°C, with 2,3-dichloropropanal as an intermediate product. They noted, however, that 2-chloroacrolein was **not** detected in this study under physiological conditions. Thus studies employing DMSO as a solvent may not be relevant for hazard assessment. 2-Chloroacrolein was found to degrade only slowly in buffer alone, but decomposed too rapidly to quantify upon addition of GSH, with or without GST.

Rat liver microsomal epoxide hydrolase and soluble epoxide hydrolase activities, when expressed in insect cells, were examined for inhibition by *cis/trans*-1,3-D epoxides of activity toward tritiated cis-stilbene oxide. When 1 mM solutions of *cis/trans*-1,3-D epoxides were incubated for various time periods prior to addition of *cis*-stilbene oxide in the presence of rat liver **microsomal** epoxide hydrolase, there was about 60% inhibition of *cis*-stilbene oxide hydrolase activity at time 0, with a decline to zero inhibition after 10 minutes. When 1 mM solutions of *cis/trans*-1,3-D epoxides were incubated for various time periods prior to addition of labeled *trans*-stilbene oxide in the presence of rat liver **soluble** epoxide hydrolase, the *trans*-stilbene oxide hydrolase activity inhibition peaked in 5 minutes at 50% inhibition, with about 35% inhibition remaining at the final sampling time of 20 minutes. Results indicate that **microsomal** epoxide hydrolase is able to hydrolyze both 1,3-epoxides within a comparatively short time. **Microsomal** epoxide hydrolase is associated with many xenobiotic responses.

A standard plate incorporation method in *S. typhimurium* TA 100, a system previously shown respond to technical Telone, found mutagenicity of 1,3-D (*cis*) to be at least 4 orders of magnitude less potent than either of the 1,3-D epoxides (*trans* 1,3-D was not tested here). A very small positive value reported for 1,3-D (*cis*) **with** S9 activation was considered to represent a treatment effect by investigators. Both epoxides elicited revertant responses, with *cis*-epoxide about 2x more potent than *trans*-epoxide. As observed in other studies, S9 had no effect on mutagenicity of either of the epoxides. For comparison, 2-chloroacrolein is about 4x more potent

than *cis*-1,3D epoxide. In contrast, 3-chloroacrolein (also not shown here to be a metabolite of 1,3-D under physiological conditions) was a very weak direct mutagen, about the same potency as *cis*-1,3-D. Varying GSH from 0 to 5 mM in TA 100 plate incorporation tests employing *cis*- and *trans*-epoxides (without GST) yielded a log-linear decrease in revertants over the entire range tested, although the *cis*-epoxide retained some mutagenicity even at the highest GSH level. Under these conditions, 2-chloroacrolein showed roughly constant mutagenicity between 0 and 0.25 mM GSH, with mutagenicity diminishing to about 25% of maximum levels at 5 mM GSH. In the latter test series, with *cis*- and *trans*-epoxides and with 2-chloroacrolein, addition of glutathione S-transferase either had no effect or decreased mutagenicity for each GSH level.

Based largely on adduct data cited in this report implicating product 3-chloro-2-hydroxypropanal as the ultimate genotoxic product, investigators proposed that "the penultimate and ultimate mutagens of 1,3-D metabolism are the corresponding epoxides and their direct hydrolysis product 3-chloro-2-hydroxypropanal, respectively." Indeed, data here show that the epoxides (or derivatives thereof) are effective mutagens, and that 3-chloro-2-hydroxypropanal (not tested in this study for mutagenicity) is a significant and comparatively persistent metabolite of the epoxides in pH 7.4 buffer.

5. Fate of 1,3-D and of 2 metabolites (3-chloroallyl alcohol and 3-chloroacrylic acid) following oral dosing in rats and mice

Bartels *et al.* (2004) (Bartels *et al.*, 2004) evaluated the fate of labeled *cis/trans* 1,3-D in male rat and mouse at an oral dose of 1 mg/kg (each species) and at 50 or 100 mg/kg respectively, and of 2 metabolic intermediates by oral dose in rats only: ^{14}C-3-chloroallyl alcohol (^{14}C-CAL) and ^{14}C-3-chloroacrylic acid (^{14}C-CAA). ^{14}C-CAL fate was examined at 5 and 65 mg/kg, whereas ^{14}C-CAA fate was examined at 5 and 75 mg/kg.

Distribution of recovered radioactivity of 1,3-D in 1 mg/kg rats was 51% in urine, 20% in feces, 18% in exhaled CO_2, and 6% in tissues. Distribution of recovered radioactivity in 1 mg/kg mice was similar: 79% in urine, 16% in feces, 14% in exhaled CO_2, and 2% in tissues. There was no apparent percent difference with either species at the higher dose level.

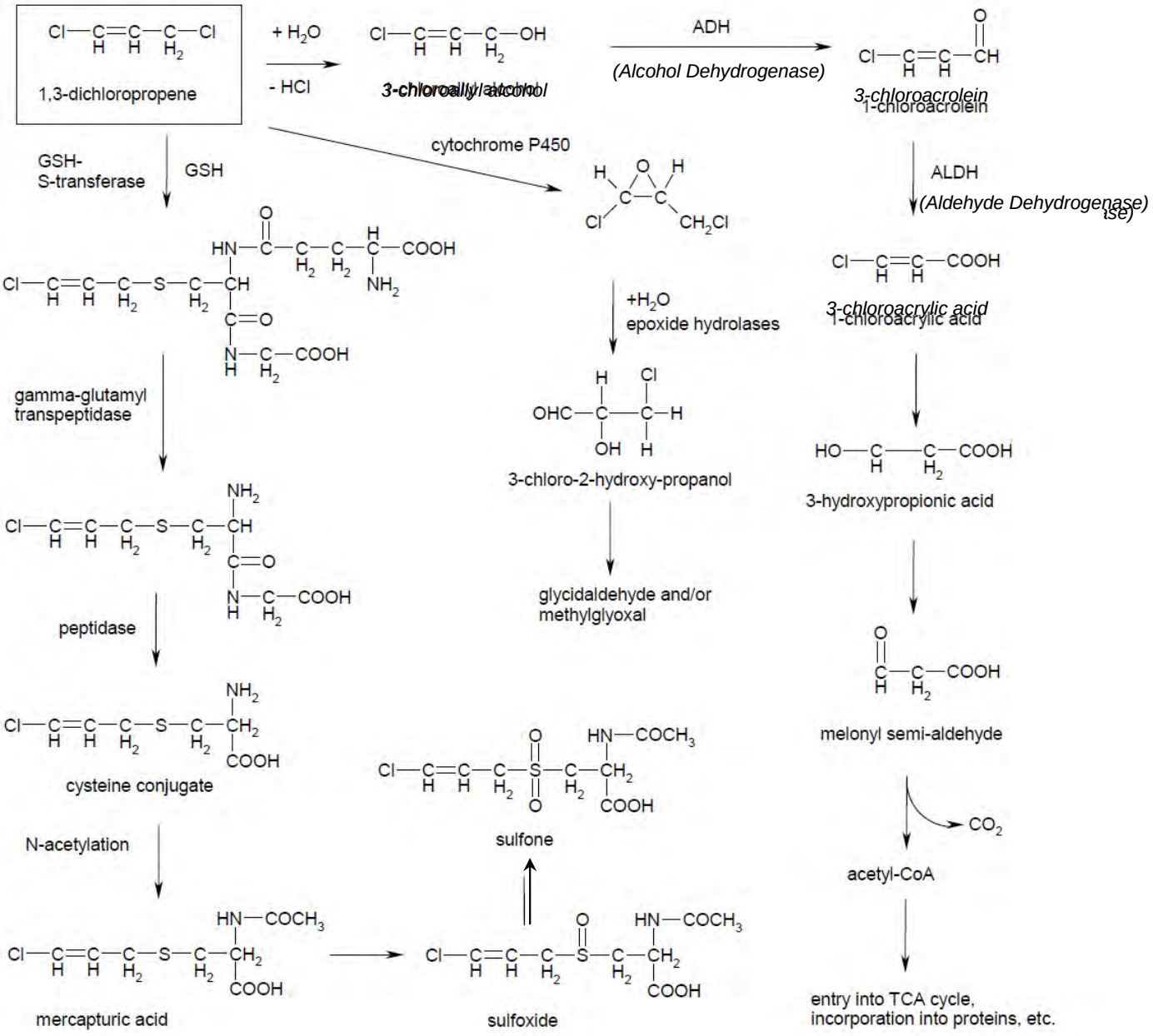
When ^{14}C-CAL was administered at 5 mg/kg, distribution of recovered radioactivity in 5 mg/kg rats was 21% in urine, 13% in feces, 52% in exhaled CO_2, and 6% in tissues. When ^{14}C-CAA was administered at 5 mg/kg, distribution of recovered radioactivity in 5 mg/kg rats was 17% in urine, 4% in feces, 68% in exhaled CO_2, and 7% in tissues. High dose distributions with both metabolites were comparable to respective low doses. Thus for both CAL and for CAA, investigators concluded that these metabolites were efficiently absorbed following oral dosing, and were both subject to substantial decomposition rather than conjugation. CAL is primarily degraded by alcohol and aldehyde dehydrogenases to CAA, with the 3-chloroacrylaldehyde (3-chloro-acrolein) as a transient (reactive) intermediate.

Investigators analyzed excreta by reverse-phase HPLC with radiochemical detection, amended by mass spectrometric analyses of structures thus separated. These investigations were performed on 1,3-D and on CAL and CAA. Profiles of the latter provided information on which major peaks derived from products of initial hydrolysis of 1,3-D on the pathway to CAL and CAA. Dominant metabolites (% of administered dose) in rats (50 mg/kg, oral) were *cis*-DCP-mercapturate (30%), *trans*-DCP-mercapturate (14%), and an assortment of co-eluting peaks containing *cis/trans*-sulfoxide and dimercapturate isomers (combined as 10%). Of the dimercapturates, the 2,3-dimercapturate was determined to derive from the epoxide via a rearrangement reaction, and the 3,3-dimercapturate derived from the route involving CAL → 3-chloro-acrolein → CAA products. Of the three major contributors to the peak containing sulfoxide and dimercapturate isomers, investigators calculated that there were roughly equal amounts of 3,3-dimercapturate and combined 1,3-D sulfoxides, with much smaller contribution by 2,3-dimercapturate (less than 2% of administered dose). Investigators determined that the fraction of absorbed dose which went through the epoxide route constitutes less than 2%, with the fraction directed through CAL and CAA a few-fold higher, with mercapturic acid products of direct glutathione conjugation being the dominant species (consistent with several other studies).

General conclusions from pharmacokinetics / metabolism studies: although most metabolism of relevant exposures of 1,3-DCP leads to relatively innocuous products, a small but toxicologically significant portion of absorbed dose is metabolized to reactive species of toxicologic concern.

Figure III.1 summarizes the metabolic fate of 1,3-D in mammals.

Figure III. 1 Metabolism of 1,3-dichloropropene in rats (adapted and updated from (ATSDR, 2008))



B. ACUTE TOXICITY

The following discussion summarizes (a) those incidents in which 1,3-D was implicated in adverse outcomes in humans under occupational, bystander and ambient exposure scenarios, and (b) all studies in which acute LC_{50} values were established in laboratory animals (and includes a table of those values (Table III.2)).

The acute / short term benchmark concentration values were derived from studies otherwise designed to determine subchronic, chronic and developmental toxicity endpoints. For this reason they are considered together in section IV (Hazard Identification), *i.e.*, after the individual study summaries.

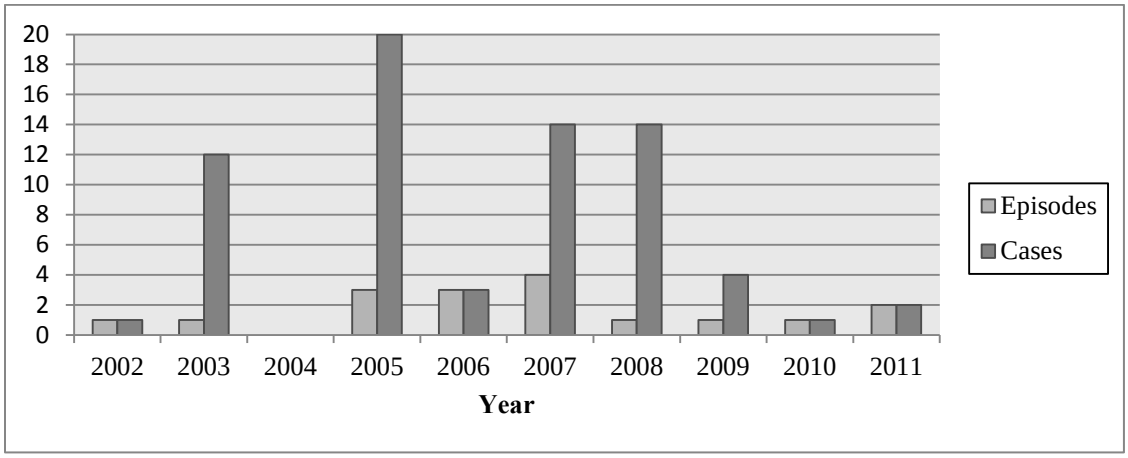
1. Illness reports in humans

In this evaluation, only DPR's Pesticide Illness Surveillance Program (PISP) database was consulted for human incident data on 1,3-dichloropropene (1,3-D). This database, though specific to California, contains all illness and injury reports potentially related to pesticide exposure and may provide patterns or trends associated with the use of a particular pesticide. PISP defines a “case” as a pesticide exposure and its apparent effects on one individual's health (CDPR, 2015b). Cases are then classified by the relationship of the exposure to a specific pesticide. A **“**definite” relationship indicates that both physical and medical evidence document exposure and subsequent health effects. A “probable” relationship indicates that limited or circumstantial evidence supports a relationship to pesticide exposure. A “possible” relationship indicates that health effects correspond generally to the reported exposure, but evidence is not available to support a relationship. PISP defines an “episode” as an incident in which one or more people experience pesticide exposure from a particular source with subsequent development or exacerbation of symptoms. Occasionally, a single episode gives rise to a large number of cases.

In California between 1982-1990, 51 cases were reported to the California Department of Pesticide Regulation’s Pesticide Illness Surveillance Program (PISP), which maintains a database of pesticide-related illnesses and injuries occurring in California (Sanborn and Powell, 1994). Case reports are received from physicians and through workers' compensation records. Of these 51 cases, the health effects attributed to exposure to 1,3-D alone, or in combination with other pesticides, were rated as definite (33 cases), probable (9 cases) or possible (9 cases). The health effects involved were systemic (16 cases), eye (14), skin (18), and combined eye-skin effects (3). From 1990 to 1995, 1,3-D use in California was suspended. As a result, from 1990 to 1997, there were no reports of illnesses associated with 1,3-D applications. In 1998, PISP identified one possible case involving 1,3-D alone with no other cases appearing until 2002. Then, in the 10 years from 2002 to 2011, the PISP identified 17 exposure episodes that gave rise to 71 cases associated with 1,3-D either alone or in combination with chloropicrin (Figure III.2) (CDPR, 2015a). The 71 cases were classified as 1 definite, 54 probable and 16 possible.

The single case reported as “definite” in 2011 included a formulation that contained both 1,3-D and chloropicrin. An employee of a soil fumigant manufacture/supplier connected a hose to an empty cylinder to purge it of any remaining chemical. The hose fell and fumigant liquid splashed into his right eye. Redness and eye irritation were reported, as well as blurred vision and burning sensation.

Figure III. 2 Numbers of illnesses (cases) and episodes reported in California, 1998-2011, evaluated by the California Pesticide Illness Surveillance Program



Two scenarios contributed to the majority of documented cases. Of the 71 cases, 64 (from 10 episodes) were due to bystander exposure, where people adjacent to recently-treated fields experienced symptoms. 6 cases were due to flushing tractor lines or repairing hoses/drip lines.

Of the 72 recently reported cases (*i.e.*, between 1998 and 2011), there were 5 cases with 1,3-D used alone between 1998 and 2011. Four of these cases exhibited respiratory symptoms. In 2007, 1 episode involving 3 cases was reported. Three mechanics were working on the air conditioning system of a fumigant tractor last used 50 days prior. The tractor was reported as dirty and each mechanic became ill after working on the tractor inside the shop. One reported chest tightness, where the other 2 reported upper respiratory tract irritation, irritated and watery eyes, dizziness, and runny nose.

The remaining 67 cases involved both 1,3-D and chloropicrin. Table III.1 summarizes the types of illnesses attributed to formulations containing both chemicals. In addition, 4 of these 67 cases involved additional pesticides including cycloate, chlorothalonil, mycoblutanil, and (most commonly) methyl bromide. Most of the cases involving 1,3-D and chloropicrin show dominance of eye effects, suggesting that the reported eye symptoms may be due to the chloropicrin.

Table III. 1 Types of Illness Cases Reported in California (1998-2011)

Illness Type [a]	Alone [b]	In Combination [c]	Total
Eye only	0	16	16
Eye & Respiratory	0	20	20
Eye, Respiratory & Systemic	2	9	1
Eye & Systemic	1	11	12
Systemic	0	2	2
Respiratory & Systemic	0	2	2
Respiratory	2	3	5
Skin	0	1	1
Other combinations of types [d]	0	3	3
Total	5	67	72

a Eye effects include irritation, burning, itching and watery eyes, and blurred vison. Respiratory illnesses include irritation of nose, throat, and lungs; coughing; chest tightness; bad taste in mouth; lung congestion; asthma and other breathing difficulties. Systemic illnesses include symptoms such as nausea, stomach ache, dizziness, headache, numbness. Skin effects include itching, and burning sensation.
b 1,3-D was applied as a sole active ingredient.
c 1,3-D formulated in a product with chloropicrin. Four cases also involved additional active ingrediates such as methyl bromide, cycloate, chlorothalonil, and mycoblutanil.
d Includes 3 less commonly combinations of eye, skin, respiratory, and systemic effects.

2. Acute LC_{50} studies on technical 1,3-D

This evaluation identified four acute or short-term LC_{50} studies on technical 1,3-D in laboratory animals. With the exception of the summary of the Kloes 1983 study, which appears in section III.G., those studies are reviewed in the following paragraphs, with the relevant LC_{50} values appearing in Table III.2. LC_{50} values for rats fell in the 600-1100 ppm range. LC_{50} values for rabbits may be lower, though this was difficult to ascertain as the only available rabbit study showed an LC_{50} of less than 300 ppm, but after exposure for 13 consecutive days. NOEL, LOEL and benchmark concentration values are summarized in Table IV.1. in section IV.A. below [1].

Streeter (1987) exposed Fischer 344 rats---5/sex/dose---to Telone II Soil Fumigant (97.5% 1,3-D) at nominal / analytical concentrations of 1076 / 1035, 946 / 855, 820 / 775 ppm for a single 4-hr period. Clinical signs included tremors, convulsions, salivation, lacrimation, diarrhea, lethargy, and other signs of altered central nervous system function. The LC_{50} for males was 855 - 1035 ppm, while that for females was 904 ppm. Converting from ppm to mg/L yielded an LC_{50} for males between 3.88 and 4.70 mg/L, and for females 4.10 mg/L. Necropsies showed facial soiling and/or hemorrhages in multiple lung lobes. Thus according to the USEPA / OPPTS Harmonized Guidelines, section 870.1, Telone II is a Toxicity Category IV pesticide for inhalation because the LC_{50} values are above 2 mg/kg.

This study was considered to be acceptable by FIFRA standards.

[1] An additional summary table containing acute and short term NOELs, LOELs and benchmark concentration values does not appear in this section because the relevant acute / short-term information came from subchronic, chronic and developmental toxicity studies. That summary table was, consequently, placed in the Hazard Identification section, *i.e.*, after the reviews of all the relevant studies. The acute LC_{50} studies were intended to address lethality, not to identify regulatory NOELs or BMCs.

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Nitschke (1990a) dosed 5 rats/sex with *cis*-1,3-dichloropropene by whole-body inhalation for 4 hours with time-weighted average concentrations of 573, 771, or 1020 ppm of *cis*-1,3-D (95.6% *cis*-1,3-D, with minor amounts of two assayed components: 1.5% *trans*-1,3-D, and 0.2% 1,2-dichloropropane). All rats died at 1020 ppm during or shortly after exposure. All males died at 771 ppm, two of which did not survive more than 30 minutes after the end of dosing. Also, 3/5 females died at 771 ppm. All rats survived at 573 ppm. Clinical signs of labored breathing and "eyelids closed" were each observed in 1/sex at 573 ppm on the day of exposure, with no clinical signs in 573 ppm rats after the day of treatment. At 573 ppm, body weights were remarkably diminished on weighing days 2 and 4, with substantial body weight recovery evident by day 8. Gross examinations revealed unilateral opacity in two high dose males, visceral congestion in four of five 771 ppm males, liver and lung congestion in all high dose males, hydrothorax in two 771 ppm males, corneal opacities in the majority of 1020 ppm females, liver and lung congestion in all high dose females, and lung edema in one mid-dose female. All 573 ppm rats were grossly normal at termination. Nominal LC_{50} was estimated to be 670 ppm and 744 ppm for males and females, respectively. For males, the most sensitive gender, mass/volume units of LC_{50} were 3.04 mg/L.

This was a supplementary study on a test article enriched in the *cis* isomer. The LC_{50} values suggest that *cis*-1,3-D is at Category IV toxicant.

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Cracknell (1987) exposed 5 Wistar-derived rats/sex/group by whole-body inhalation for 4 hours with Telone II (98.4%) at 0, 1.62, 2.64, 2.70, or 3.07 mg/L (assayed mean concentrations). Rats were observed at 6 intervals during exposure, and daily during a 14-day post-dose observation period. All rats survived except for 3/5 males and 3/5 females at 3.07 mg/L. All male groups showed a body weight loss at the day 1 weighing, followed by normal subsequent gains in survivors. Body weights were unaffected in 1.62 mg/L females, but other female groups suffered body weight losses on day 1, followed by normal weight gain patterns thereafter. An exception was that the surviving 3.07 mg/L females appeared to require an additional 5 days for weight gains to normalize. Common signs at the lowest dose (1.62 mg/L) during exposure were partial closing of eyes, slow respiratory rate and irregular respiratory movements during exposure in all rats; as well as hunched posture, restlessness, exaggerated respiratory movements, and pawing behavior in some rats. Clinical signs for 1.62 mg/L were normal by the day after treatment, whereas symptoms continued for up to 5 days at 3.07 mg/L. LC_{50} was thus between 2.70 (585 ppm) and 3.07 (665 ppm) mg/L in both sexes.

While the study was not considered to be acceptable by FIFRA standards, it was upgraded by DPR upon submission of a standard curve and sample calculation to validate the reported assayed test atmosphere content.

Table III. 2 LC_{50} values resulting from acute or short-term inhalation exposure to 1,3-D in rats and rabbits

Test article	Species	LC_{50}	Toxicity Category	Reference
Telone II (97.5% 1,3-D)	Fischer 344 rats / 4 hr	855-1035 ppm (♂) 904 ppm (♀)	IV	(Streeter *et al.*, 1987)
***Cis*-1,3-D**	Fischer 344 rats / 4 hr	670 ppm (♂) 744 ppm (♀)	IV	(Nitschke *et al.*, 1990a)
Telone II (98.4% 1,3-D)	Wistar rats / 4 hr	585-665 ppm (♂ & ♀)	IV	(Cracknell *et al.*, 1987)
Telone II (92.1% 1,3-D) [a]	Pregnant Fischer 344 rats / 6 hr/d, gd 6-15	>300 ppm (♀): 1 died	II	(Kloes *et al.*, 1983) [b]
Telone II (92.1% 1,3-D) [a]	Pregnant New Zealand White rabbits / 6 hr/d, gd 6-18	<300 ppm (♀): 5/6 died	II	(Kloes *et al.*, 1983) [b]

[a] Contains 1% epichlorohydrin

[b] This study is summarized in section III.G. (Developmental Toxicity) below.

C. SUBCHRONIC TOXICITY

1. Rat

Stott (1984) exposed Fischer 344 rats and B6C3F1 mice (10/sex/dose, both species) to Telone II (90.9% 1,3-D) for 13 weeks: 6 hr/day, 5 days/wk. This summary concerns only the rat data. The target concentrations were 0, 10, 30, 90 and 150 ppm, which were equivalent to 9.1, 27.3, 81.8 and 136 ppm 1,3-D when the test article purity was taken into account. As with the chronic studies, a J-tube apparatus was used to vaporize the test article and meter it into the chambers. 1,3-D was monitored 2-3x/hr by infrared spectrophotometry. Analytical concentrations were within 5% of the target concentrations. The range of Telone distribution within the chamber was within 10%.

Twice daily observations were carried out on exposure days (*i.e.*, 5 days/wk) for mortality, moribundity and clinical signs. Body weights were determined weekly. Food consumption was not monitored. Hematology, clinical chemistry and urinanalyses were performed at the end of the study. Gross necropsies were conducted on fasted survivors. Histopathology was done on representative sections of major organs and tissues from both species. Particular attention was paid to nervous tissues, where a series of special stains were used.

Mortality, moribundity and clinical signs. Telone II had no discernable effect on mortality. There were no Telone-associated clinical signs.

Body weights. Mean rat body weights were statistically reduced by the time of the first measurement at 3 days at 90 and 150 ppm in both males and females (Table III.3). Body weight deficits compared to controls were 5-9% and 5-8% at 90 ppm in males and females, respectively. At 150 ppm the deficits were 9-20% and 9-19%. A possible effect was also noted in females at 30 ppm (0-4% deficit compared to controls), though statistical significance was not achieved at that concentration.

Organ weights. Statistically significant changes in the following organ weights were noted in brain, heart, kidney, liver and testicle at 90 and 150 ppm in both sexes (Table III.3). The report considered these findings to be “consistent with the proportionately lower body fat content and nonparenchymal cell mass of these animals…” (p. 12). As there were no histopathologic correlates to these changes, they were interpreted by the investigators as “an indirect, nonspecific result of Telone II vapor exposure”.

Clinical chemistry, hematology and urinalysis. Statistically reduced serum protein levels at 90 (females) and 150 (males and females) ppm and slightly increased RBC counts (females) were observed at 30, 90 and 150 ppm. These were attributed to poor nutrition, which in turn may have resulted from reduced food intake (not measured). As no parallel histopathologic changes were noted, these effects were not accorded toxicologic significance. Urinalysis did not show significant changes.

Gross pathology and histopathology. Only one effect attributable to Telone was observed at the gross pathological level in rats: females showed decreased abdominal fat at 150 ppm (0/10, 0/10, 0/10, 0/10, 7/10). This was almost certainly related to the reduced body weights noted at that dose. Histological analysis revealed the following lesions at the high dose: slight degeneration of the nasal olfactory epithelium, slight hyperplasia of the nasal respiratory epithelium, incomplete development of the uterus (slight hypoplasia) and mesenteric adipose atrophy in females (Table III.4). Very slight hyperplasia of the nasal respiratory epithelium was also noted at 30 and 90 ppm.

The NOEL was set at 10 ppm based on hyperplasia of the nasal respiratory epithelium in 2/10 males at 30 ppm. It should be noted that body weight decreases were noted by day 3 in both sexes, and thus may be pertinent to the evaluation of potential risks arising from acute or short term exposures. The study was considered to be supplemental.

Table III. 3 Effect of inhalation exposure to Telone on body and organ weight in Fischer 344 rats over a 13-wk period (Stott *et al.*, 1984)

Males

	Telone, ppm				
	0	**10**	**30**	**90**	**150**
Body weights (grams)					
Day -1	176.0±9.1	180.3±6.0	177.2±4.9	175.6±7.6	175.9±8.5
3	191.6±11.3	194.8±7.2	190.1±7.2	181.1±7.9*	174.3±9.6*
11	208.9±11.8	210.8±8.1	206.4±8.4	192.1±9.1*	184.1±10.1*
18	221.4±14.3	223.2±10.9	222.2±10.3	201.2±9.8*	190.7±10.7*
25	234.0±16.5	235.4±12.2	234.2±10.6	213.3±8.5*	199.1±10.6*
32	242.9±19.6	252.3±12.9	249.0±12.1	227.7±8.3	210.3±12.9*
39	259.5±20.1	266.9±12.0	260.5±11.0	238.9±8.0*	217.7±13.4*
46	272.3±20.7	280.2±11.2	272.1±12.6	248.7±9.3*	228.4±14.7*
53	279.1±21.6	289.7±12.8	282.5±14.3	256.7±9.4*	233.3±13.6*
60	286.5±23.0	295.1±13.7	286.8±12.9	262.8±9.6*	236.9±13.2*
67	296.4±22.5	306.5±14.9	296.2±14.8	271.0±8.9*	244.3±14.6*
74	303.6±21.6	313.5±15.6	302.1±14.2	275.6±8.6*	246.1±14.8*
81	307.4±21.5	320.0±16.4	309.8±16.5	280.4±7.7*	250.4±14.5*
88	308.3±21.8	321.1±15.9	310.0±17.5	282.2±8.8*	247.9±14.7*
Organ weights (absolute & relative to body weight)					
Brain					
■ grams	1.884±0.056	1.931±0.034	1.918±0.048	1.907±0.042	1.827±0.056*
■ grams/100 g	0.659±0.035	0.647±0.028	0.668±0.031	0.727±0.025*	0.785±0.037*
Heart					
■ grams	0.830±0.051	0.872±0.035	0.846±0.042	0.817±0.031	0.754±0.056*
■ grams/100 g	0.289±0.007	0.292±0.009	0.294±0.011	0.311±0.012*	0.323±0.016*
Kidney					
■ grams	2.051±0.202	2.101±0.114	2.067±0.136	1.988±0.133	1.874±0.132*
■ grams/100 g	0.714±0.034	0.702±0.013	0.718±0.028	0.757±0.044*	0.804±0.038*
Liver					
■ grams	7.318±0.598	7.765±0.388	7.414±0.619	7.119±0.384	6.611±0.445*
■ grams/100 g	2.551±0.108	2.595±0.052	2.574±0.108	2.711±0.104*	2.835±0.114*
Testicle					
■ grams	3.071±0.267	3.225±0.138	3.186±0.210	3.166±0.106	3.082±0.139*
■ grams/100 g	1.074±0.101	1.079±0.016	1.109±0.071	1.207±0.057*	1.323±0.054*
Thymus					
■ grams	0.240±0.028	0.261±0.040	0.247±0.023	0.234±0.021	0.205±0.044
■ grams/100 g	0.084±0.006	0.087±0.013	0.086±0.007	0.089±0.009	0.087±0.017

* α=0.05, Dunnett's test (two-sided)

Table III. 3 (continued). Effect of inhalation exposure to Telone on body and organ weight in Fischer 344 rats over a 13-wk period (Stott *et al.*, 1984)

Females

Telone, ppm					
Day	**0**	**10**	**30**	**90**	**150**
Body weights (grams)					
-1	131.7±6.0	131.7±5.3	131.2±5.9	130.6±4.1	131.2±5.5
3	138.9±6.4	137.5±5.1	138.5±5.7	131.5±5.3*	126.9±6.2*
11	143.0±7.8	142.8±4.7	141.2±5.8	135.3±5.4*	130.6±5.2*
18	149.1±7.9	148.8±4.4	146.1±5.0	139.2±5.8*	133.8±6.7*
25	152.5±9.4	151.6±4.3	148.7±4.8	142.2±6.3*	137.9±8.3*
32	157.3±8.9	158.7±4.2	154.6±4.8	148.2±7.6*	142.5±9.7*
39	161.7±8.8	163.6±4.5	157.1±5.1	151.5±6.4*	140.5±11.3*
46	165.4±9.0	167.8±6.1	163.3±6.4	155.6±7.8*	144.4±11.9*
53	168.5±8.9	170.7±5.6	166.5±6.1	158.3±5.4*	146.4±12.0*
60	171.2±8.8	171.8±6.3	168.4±5.6	160.8±6.6*	146.7±13.8*
67	175.1±8.4	175.4±6.9	170.0±6.7	163.6±6.8*	147.4±15.0*
74	176.6±9.0	176.9±6.3	171.0±6.1	164.1±7.0[@]	146.9±16.6[@]
81	180.1±7.3	178.2±7.4	172.6±6.7	165.2±7.0[@]	146.6±17.3[@]
88	175.1±10.2	174.8±7.8	170.8±6.8	164.3±6.6[@]	143.6±18.0[@]
Organ weights (absolute & relative to body weight)					
Brain					
■ grams	1.717±0.046	1.697±0.065	1.703±0.038	1.695±0.043	1.619±0.083*
■ grams/100 g	1.065±0.048	1.048±0.028	1.080±0.027	1.114±0.039	1.223±0.114*
Heart					
■ grams	0.554±0.035	0.544±0.028	0.531±0.036	0.526±0.020	0.491±0.026*
■ grams/100 g	0.343±0.018	0.336±0.014	0.337±0.015	0.346±0.015	0.370±0.032*
Kidney					
■ grams	1.261±0.127	1.261±0.060	1.236±0.084	1.290±0.074	1.199±0.085
■ grams/100 g	0.781±0.065	0.780±0.039	0.783±0.032	0.847±0.042*	0.904±0.065*
Liver					
■ grams	4.251±0.395	4.276±0.158	4.130±0.258	4.151±0.257	3.646±0.362*
■ grams/100 g	2.633±0.204	2.643±0.078	2.617±0.114	2.725±0.077	2.737±0.114
Testicle					
■ grams	n/a				
■ grams/100 g					
Thymus					
■ grams	0.192±0.027	0.191±0.028	0.183±0.023	0.187±0.021	0.137±0.045*
■ grams/100 g	0.119±0.014	0.118±0.015	0.116±0.016	0.123±0.014	0.100±0.024

* α=0.05, Dunnett's test (two-sided)

Table III. 4 Histopathologic observations in Fischer 344 rats following 13 weeks of inhalation exposure to Telone (Stott *et al.*, 1984)

	Telone (ppm)									
	Males					Females				
	0	10	30	90	150	0	10	30	90	150
Nasal turbinates										
■ hyperplasia, respiratory epithelium, very slight	0/10	0/10	2/10	10/10	0/10	0/10	0/10	0/10	10/10	0/10
■ hyperplasia, respiratory epithelium, slight	0/10	0/10	0/10	0/10	10/10	0/10	0/10	0/10	0/10	10/10
■ degeneration, olfactory epithelium, slight	0/10	0/10	0/10	0/10	10/10	0/10	0/10	0/10	0/10	0/10
Uterus										
■ hypoplasia, slight	n/a	n/a	n/a	n/a	n/a	0/10	0/10	0/10	0/10	7/10
Mesenteric tissue										
■ adipose atrophy	0/10	nd	nd	nd	0/10	1/10	nd	nd	nd	7/10

2. Mouse

Stott (1984) exposed Fischer 344 rats and B6C3F1 mice (10/sex/dose, both species) to Telone II (90.9% 1,3-D) for 13 weeks: 6 hr/day, 5 days/wk. *This summary concerns only the mouse data*. The target concentrations were 0, 10, 30, 90 and 150 ppm, which were equivalent to 9.1, 27.3, 81.8 and 136 ppm 1,3-D when the test article purity was taken into account. As with the chronic studies, a J-tube apparatus was used to vaporize the test article and meter it into the chambers. 1,3-D was monitored 2-3x/hr by infrared spectrophotometry. Analytical concentrations were within 5% of the target concentrations. The range of Telone distribution within the chamber was within 10%.

Twice daily observations were carried out on exposure days (*i.e.*, 5 days/wk) for mortality, moribundity and clinical signs. Body weights were determined weekly. Food consumption was not monitored. Hematology and clinical chemistry were performed at the end of the study (urinalyses were not performed). Gross necropsies were conducted on fasted survivors. Histopathology was done on representative sections of major organs and tissues from both species.

Mortality, moribundity and clinical signs. Three mice suffered spontaneous deaths: a control female on day 3 (handling trauma), a 90-ppm female on day 3 (broken back due to handling trauma) and a 150-ppm female (undetermined cause). One control male was sacrificed moribund on day 31 (broken back). None of the deaths were attributed to Telone exposure, nor were there Telone-associated clinical signs.

Body weights. Mean mouse body weights were statistically reduced at 150 ppm in both sexes starting from day 3 (2-14% in males, 4-13% in females). Body weights were also generally lower at 90 ppm (0-6% in males, 3-12% in females), but statistical significance was present only rarely (Table III.5).

Organ weights. Statistically significant changes in organ weights were noted in brain, heart, kidney, liver and thymus at 90 and 150 ppm in both sexes (Table III.5). These were considered to be secondary expressions of the reduced weight gains at those doses. Also, as noted by the investigators, "…the changes noted in the kidney weights of female mice may have been related in some way to the observed effects of Telone II upon the urinary bladder epithelium of these animals…" (p. 13).

Clinical chemistry and hematology. Serum blood urea nitrogen levels were reduced in a dose-dependent manner in males, with statistical significance achieved at 90 and 150 ppm (at ascending doses, mg/dl: 34±7, 30±7, 26±6, 20±4*, 15±1*; α=0.05, two-sided). The investigators attributed this to depressed body weights and moderate food deprivation. SGPT (serum glutamic-pyruvic transaminase) levels were increased in both sexes, achieving significance in females (males, mU/ml: 66±54, 43±17, 58±33, 43±15, 117±78; females: 38±11, 35±16, 39±34, 48±31, 70±23*; α=0.05, two-sided). However, the toxicologic significance of this observation was unclear in light of the absence of histopathologic impacts on the liver.

Gross pathology and histopathology. Gross pathology in mice revealed only one sign: decreased male thymus size at 150 ppm (1/9, 0/10, 0/10, 0/10, 9/10), which was also reflected in the decreased thymus weights noted in Table III.5. Histopathology revealed slight degeneration of the olfactory neuroepithelium and a slight hyperplasia of the respiratory epithelium in both sexes at 90 and 150 ppm (Table III.6). These lesions were accompanied by focal respiratory metaplasia ("a condition in which the damaged sensory olfactory epithelium is replaced by ciliated respiratory epithelium identical to that lining the remainder of the nasal cavity and respiratory tract"; p. 17). Moderate hyperplasia of the urinary bladder transitional epithelium was also noted in 90 and 150 ppm females, with mild aggregations of lymphoid cells in subepithelial tissues in some of these mice at as low as 30 ppm.

The NOEL was set at 30 ppm based on body weight decrements, organ weight changes, nasal histopathology and urinary bladder histopathology at 90 and 150 ppm. The early body weight changes were considered pertinent to the establishment of a short term point of departure. For calculation of the appropriate Human Equivalent Concentrations, see section IV.F below.

This study was considered to be supplemental.

Table III. 5 Effect of inhalation exposure to Telone on body weight in B6C3F1 mice over a 13-wk period (Stott *et al.*, 1984)

Males

Telone, ppm					
Day	**0**	**10**	**30**	**90**	**150**
Body weights (grams)					
-1	24.6±0.8	25.1±1.1	24.3±1.2	24.9±1.2	24.0±0.8
3	24.7±1.4	25.4±1.2	24.4±1.2	25.2±1.8	24.1±1.0
10	26.2±1.2	27.6±1.4	26.7±1.0	26.6±1.5	24.9±1.5
17	26.9±1.3	28.1±1.8	27.2±1.2	26.3±1.6	24.5±1.1*
24	26.7±1.0	27.9±1.5	26.8±0.8	26.1±1.0	24.5±1.2*
31	28.2±1.3	28.6±1.6	27.7±1.2	27.1±1.4	24.9±1.5*
38	28.2±1.0	29.4±1.5	28.4±1.2	27.2±1.3	25.1±1.6*
45	28.8±1.1	29.0±1.5	28.7±1.2	27.4±1.1	25.2±1.1*
52	27.8±0.7	28.6±1.8	28.3±1.1	27.1±1.2	25.3±1.2*
59	29.2±1.2	29.5±1.3	29.1±1.2	27.4±1.6*	25.7±1.1*
66	29.6±1.1	30.3±1.3	29.7±1.3	28.7±1.6	25.6±1.1*
73	30.0±1.3	30.4±1.6	29.9±1.2	28.9±1.7	26.3±1.1*
80	30.3±1.1	31.1±1.6	30.7±1.5	29.0±1.5	26.4±1.0*
87	30.5±1.5	31.3±1.6	30.5±1.6	29.5±1.7	26.7±1.1*
Organ weights (absolute & relative to body weight)					
Brain					
■ grams	0.467±0.015	0.464±0.017	0.460±0.014	0.454±0.010	0.443±0.017*
■ grams/100 g	1.608±0.053	1.581±0.070	1.561±0.084	1.602±0.073	1.721±0.065*
Heart					
■ grams	0.148±0.013	0.140±0.015	0.141±0.016	0.130±0.009*	0.113±0.008*
■ grams/100 g	0.508±0.036	0.476±0.041	0.477±0.046	0.459±0.027*	0.439±0.028*
Kidney					
■ grams	0.471±0.022	0.492±0.048	0.481±0.040	0.447±0.025	0.391±0.023*
■ grams/100 g	1.621±0.075	1.670±0.119	1.629±0.104	1.575±0.036	1.516±0.061*
Liver					
■ grams	1.673±0.113	1.656±0.128	1.598±0.140	1.431±0.184*	1.271±0.087*
■ grams/100 g	5.756±0.316	5.629±0.331	5.405±0.327	5.038±0.537*	4.937±0.374*
Testicle					
■ grams	0.225±0.017	0.218±0.033	0.225±0.018	0.217±0.021	0.211±0.020
■ grams/100 g	0.774±0.069	0.741±0.109	0.762±0.052	0.764±0.070	0.818±0.075
Thymus					
■ grams	0.036±0.010	0.040±0.006	0.042±0.005	0.029±0.005	0.026±0.006*
■ grams/100 g	0.124±0.034	0.135±0.018	0.141±0.017	0.101±0.018	0.101±0.025

* α=0.05, Dunnett's test (two-sided)

Table III. 5 (continued) Effect of inhalation exposure to Telone on body weight in B6C3F1 mice over a 13-wk period (Stott *et al.*, 1984)

Females

Telone, ppm					
Day	**0**	**10**	**30**	**90**	**150**
Body weights (grams)					
-1	18.9±0.7	18.8±1.0	19.2±0.7	19.0±0.8	18.4±1.1
3	19.8±1.3	19.6±1.2	19.4±0.6	19.0±1.0	18.1±1.2*
10	20.4±0.7	20.5±1.1	20.9±0.7	19.9±1.1	19.6±0.9
17	21.5±0.9	21.6±1.2	21.3±0.5	20.8±1.4	19.5±1.1*
24	21.6±0.8	21.9±0.9	22.3±1.4	20.4±1.5	19.4±1.4*
31	22.6±1.1	21.7±1.5	22.4±0.6	21.4±1.5	20.5±1.4*
38	22.9±0.9	23.1±1.1	23.5±0.7	21.5±1.5	21.1±1.7*
45	23.8±1.0	23.5±1.1	24.3±0.6	21.9±1.7*	20.8±1.2*
52	24.0±1.0	23.2±1.2	23.5±0.5	23.0±20.	21.0±1.3*
59	24.1±0.6	23.9±1.3	24.3±0.5	22.6±1.6[@]	21.3±1.7[@]
66	24.0±1.1	24.3±1.1	24.9±1.1	23.4±1.7	21.9±1.5*
73	25.0±0.7	24.5±1.3	24.6±1.0	23.8±1.9	22.3±1.4*
80	25.4±0.8	24.7±1.4	25.0±0.7	24.0±1.7	22.8±1.3*
87	25.5±0.8	25.8±1.2	25.9±0.8	24.7±2.0	23.0±1.3*
Organ weights (absolute & relative to body weight)					
Brain					
■ grams	0.466±0.016	0.457±0.014	0.464±0.032	0.449±0.019	0.425±0.021*
■ grams/100 g	1.861±0.053	1.833±0.094	1.858±0.136	1.850±0.111	1.876±0.073
Heart					
■ grams	0.113±0.009	0.112±0.009	0.115±0.009	0.109±0.013	0.099±0.011*
■ grams/100 g	0.453±0.035	0.450±0.035	0.462±0.043	0.446±0.034	0.436±0.026
Kidney					
■ grams	0.327±0.028	0.325±0.031	0.330±0.022	0.350±0.039	0.324±0.040
■ grams/100 g	1.305±0.074	1.301±0.079	0.322±0.073	1.437±0.125*	1.423±0.102
Liver					
■ grams	1.430±0.133	1.392±0.133	1.370±0.118	1.312±0.121	1.154±0.147*
■ grams/100 g	5.699±0.305	5.569±0.381	5.475±0.366	5.394±0.355	5.069±0.390*
Testicle					
■ grams	n/a				
■ grams/100 g					
Thymus					
■ grams	0.053±0.008	0.47±0.009	0.045±0.009	0.040±0.007*	0.031±0.006*
■ grams/100 g	0.212±0.033	0.187±0.039	0.180±0.037	0.165±0.024*	0.138±0.025*

* α=0.05, Dunnett's test (two-sided)

[@] α=0.05, Wilcoxon's test (two-sided)

Table III. 6 Histopathologic observations in B6C3F1 mice following 13 weeks of inhalation exposure to Telone (Stott *et al.*, 1984)

	Telone (ppm)									
	Males					Females				
	0	10	30	90	150	0	10	30	90	150
Nasal turbinates										
■ degeneration, olfactory epithelium, very slight	0/9	0/10	0/10	10/10	0/10	0/9	0/10	0/10	9/10	0/9
■ degeneration, olfactory epithelium, slight	0/9	0/10	0/10	0/10	10/10	0/9	0/10	0/10	0/9	9/9
■ hyperplasia, respiratory epithelium, very slight	0/9	0/10	0/10	10/10	1/10	0/9	0/10	0/10	9/9	2/9
■ hyperplasia, respiratory epithelium, slight	0/9	0/10	0/10	0/10	9/10	0/9	0/10	0/10	0/9	7/9
■ metaplasia, olfactory epithelium, multifocal, slight	0/9	0/10	0/10	0/10	10/10	0/9	0/10	0/10	0/9	6/9
Urinary bladder										
■ hyperplasia, epithelial cells, moderate	0/9	0/10	0/10	0/10	0/10	0/9	0/10	0/10	7/9	6/9
■ aggregates of mononuclear cells, submucosa	2/9	1/10	0/10	2/10	0/10	2/9	3/10	9/10	6/9	4/9

3. Rat and mouse

Fischer 344 rats and CD-1 albino mice---10/sex/dose for both species---were subjected to whole-body exposure to Telone II ("production grade", purity not stated) for 6 hr/day, 5 days/wk, for 13 weeks (Coate, 1979). Telone vapors were generated from the liquid formulation by heating followed by metering the resultant vapor through a manifold in which rates of air flow were controlled to create different concentrations in the exposure chambers. The target 1,3-D concentrations were 0, 10, 30 and 90 ppm. Mean measured concentrations were 0, 11.98±4.01, 32.14±8.41 and 93.02±25.93 ppm. Animals were observed for clinical signs both before and after each exposure period and at each weighing. Body weights were determined at the outset of the study and weekly thereafter until sacrifice, which occurred within 48 hours of the final exposure. Complete gross necropsies and histopathology were carried out on a range of tissues from both species.

Rats. Neither deaths nor 1,3-D related clinical signs were observed throughout the study. Statistically significant body weight reductions were noted particularly in high dose males during

weeks 6-13 and in high dose females throughout the study (Table III.7). Gross necropsies did not reveal treatment-related effects. Histopathology was negative except for the appearance of decreased cytoplasm and nuclear disorganization in the epithelial lining of the nasal septum and dorsal turbinates at the mid and high doses in females and at the high dose in males, and necrosis of individual nasal cells at the high dose in both sexes (Table III.8). Response intensity increased at the high dose. The rat NOEL was 12 ppm based on the nasal histopathologic effects in females at the LOEL of 32 ppm.

Mice. There were no treatment-related deaths during the study, though two low-dose males died from fighting or accident-related injuries. Statistically significant body weight reductions were noted in high dose females throughout the study (Table III.9). Males did not show a clear effect of treatment on body weight gain at any dose. Except for the 3/10 high dose males with dark renal medulla of uncertain toxicologic significance, gross necropsies did not reveal treatment-related effects. Histopathology was negative except for the appearance of decreased cytoplasmic content in the epithelial cells of the nasal septum and dorsal turbinates of 6/10 high dose females and necrosis of individual nasal septum cells in 1/10 females (Table III.10). The mouse NOEL was set at 32 ppm based on nasal histopathology in females at the LOEL of 93 ppm.

This study was considered supplemental because it was intended as a limited pilot study to test the ability of rats and mice to survive much longer exposures. However, the data were considered adequate for NOEL and LOEL designations.

Table III. 7 Effect of inhalation exposure to Telone II on body weight in F344 rats over a 13-wk period (Coate, 1979)

Week	0 ppm	10 ppm	30 ppm	90 ppm
		Males (grams)		
0	173±8.2	167±11.2	176±16.4	172±18.2
1	192±9.9	180±5.9	190±14.8	187±10.2
2	204±8.6	211±5.5	197±10.7*	194±12.9*
3	198±8.0	217±11.7	216±25.1	210±15.2
4	232±7.4	237±7.1	235±12.2	223±19.5
5	243±8.4	252±7.4	247±13.2	234±18.5
6	255±7.5	261±8.3	257±13.4	243±20.4*
7	262±9.4	267±8.5	259±15.6	247±24.7*
8	267±9.0	277±9.2	270±14.6	247±21.4*
9	276±10.2	283±10.8	275±15.3	258±21.2*
10	204±13.0	289±11.3	285±12.8	258±20.3*
11	291±12.8	295±11.8	291±13.6	271±21.9*
12	298±15.6	299±12.9	297±13.2	273±24.4*
13	301±17.3	303±13.7	301±12.6	278±24.1*
		Females (grams)		
0	134±6.8	136±6.9	132±2.5	133±6.2
1	149±7.4	146±5.5*	141±6.9*	141±5.0*
2	152±9.9	152±6.5	149±5.5	140±6.3*
3	163±7.0	163±12.4	164±5.0	152±6.3*
4	169±7.3	170±8.4	164±7.4	154±5.8*
5	171±6.7	173±8.2	168±5.0	157±6.1*
6	173±7.6	176±6.9	175±3.9	161±7.1*
7	176±8.0	180±9.6	177±5.0	163±5.7*
8	181±8.6	185±8.9	179±3.9	166±6.2*
9	180±9.1	188±7.1	184±4.0	168±5.7*
10	188±9.4	190±8.8	185±5.1	171±5.6*
11	191±8.6	193±8.5	189±4.0	172±7.5*
12	194±8.2	194±9.1	188±4.2	178±7.1*
13	194±9.1	196±8.4	190±4.5	178±6.5*

Note: measured 1,3-D concentrations were 0, 11.98±4.01, 32.14±8.41 and 93.02±25.93 ppm.

* α=0.05, one-way classification for covariance (each animal's pre-exposure body weight as covariate)

Table III. 8 Effect of inhalation exposure to Telone II on nasal histopathology in F344 rats over a 13-wk period (Coate, 1979)

	Males				Females			
	0 ppm	**10 ppm**	**30 ppm**	**90 ppm**	**0 ppm**	**10 ppm**	**30 ppm**	**90 ppm**
Decreased cytoplasm	0/10	0/10	0/10	10/10	0/10	0/10	9/10	10/10
Disorganization of nuclei	0/10	0/10	0/10	10/10	0/10	0/10	8/10	10/10
Necrosis of individual cells	0/10	0/10	0/10	3/10	0/10	0/10	0/10	4/10

Note: measured 1,3-D concentrations were 0, 11.98±4.01, 32.14±8.41 and 93.02±25.93 ppm.

Table III. 9 Effect of inhalation exposure to Telone II on body weight in CD-1 mice over a 13-wk period (Coate, 1979)

Week	0 ppm	10 ppm	30 ppm	90 ppm
		Males (grams)		
0	38.7±3.51	41.7±4.5	40.2±2.97	39.0±2.06
1	40.2±3.00	41.6±3.63	41.3±2.47	40.6±2.03
2	42.1±3.74	43.4±3.99	42.1±2.92	40.8±2.21
3	39.6±4.00	43.8±3.72	44.0±2.92	40.9±2.81
4	42.5±3.98	44.4±4.5	44.3±3.42	41.3±2.32
5	44.4±4.17	43.6±5.12	45.8±3.56	41.9±2.88
6	44.8±4.32	47.0±5.24	44.7±3.71	43.1±2.63
7	45.8±4.33	46.4±7.00	45.9±3.57	43.2±2.51
8	47.4±4.62	47.6±4.07	47.3±3.81	43.8±2.69*
9	47.5±4.86	48.8±4.80	48.0±4.49	44.0±3.04
10	47.0±4.44	50.5±5.18	47.3±4.22	44.5±3.26
11	43.0±5.66	48.8±4.47	47.3±3.70	44.7±3.13
12	48.4±4.32	49.6±4.11	47.5±4.26	45.1±3.20
13	49.1±4.50	50.7±4.44	47.8±4.87	45.4±3.31
		Females (grams)		
0	29.1±1.47	29.6±2.14	29.4±1.88	29.2±2.36
1	30.9±1.14	30.9±1.74	31.2±2.34	29.6±2.50*
2	31.3±1.55	32.3±2.44	32.6±2.51	29.0±2.15*
3	33.0±1.49	32.5±2.03	32.8±2.00	30.3±2.65*
4	33.6±1.58	34.0±2.46	34.4±2.11	30.8±2.66*
5	34.3±1.48	33.5±2.97	34.2±1.84	30.8±2.38*
6	34.0±1.52	34.2±2.49	34.7±1.86	31.3±2.47*
7	34.6±1.78	34.7±3.06	34.6±2.31	31.7±2.23*
8	36.0±1.57	35.9±2.89	36.9±2.45	32.7±2.64*
9	35.6±2.02	34.5±2.99	36.0±2.14	30.8±2.21*
10	36.2±2.46	36.5±3.47	37.6±2.21	32.6±2.55*
11	37.8±2.34	36.8±3.30	37.6±2.36	33.1±2.49*
12	38.4±2.34	36.9±3.02	38.0±2.81	34.3±2.40*
13	38.1±2.15	37.4±3.40	37.5±2.85	34.0±2.57*

Note: measured 1,3-D concentrations were 0, 11.98±4.01, 32.14±8.41 and 93.02±25.93 ppm.

* α=0.05, one-way classification for covariance (each animal's pre-exposure body weight as covariate)

Table III. 10 Effect of inhalation exposure to Telone II on nasal histopathology in F344 rats over a 13-wk period (Coate, 1979)

	Males				Females			
	0 ppm	**10 ppm**	**30 ppm**	**90 ppm**	**0 ppm**	**10 ppm**	**30 ppm**	**90 ppm**
Decreased cytoplasm	0/10	0/10	0/10	0/10	0/10	0/10	0/10	6/10
Necrosis of individual cells	0/10	0/10	0/10	0/10	0/10	0/10	0/10	1/10

Note: measured 1,3-D concentrations were 0, 11.98±4.01, 32.14±8.41 and 93.02±25.93 ppm.

Table III. 11 Summary of NOEL and LOEL values from subchronic inhalation studies on 1,3-D

Study type	NOEL	LOEL	Determining sign(s)	*Acceptability* Reference
■ Rat inhalation ■ 6 hr/day, 5 d/wk, 13 wk ■ 0, 10, 30, 90 & 150 ppm	10 ppm	30 ppm [a]	Hyperplasia of nasal respiratory epithelium	*Supplemental* (Stott *et al.*, 1984)
■ Rat inhalation ■ 6 hr/day, 5 d/wk, 13 wk ■ 0, 10, 30, & 90 ppm	12 ppm	32 ppm	Nasal histopathology in females	*Supplemental* (Coate, 1979)
■ Mouse inhalation ■ 6 hr/day, 5 d/wk, 13 wk ■ 0, 10, 30, 90 & 150 ppm	30 ppm	90 ppm	Body weight decrements, organ weight changes, nasal histopathology and urinary bladder histopathology	*Supplemental* (Stott *et al.*, 1984)
■ Mouse inhalation ■ 6 hr/day, 5 d/wk, 13 wk ■ 0, 10, 30, & 90 ppm	32 ppm	93 ppm	Nasal histopathology in females	*Supplemental* (Coate, 1979)

[a] Critical LOEL value. BMCL modeling of the nasal histopathology incidence data resulted in a critical $BMCL_{10}$ of 16 ppm for this study (Appendix III).

D. CHRONIC TOXICITY AND ONCOGENICITY

1. Rat inhalation

Lomax (1987) administered Telone II soil fumigant [2] by whole-body inhalation to F344 rats, 6-8 weeks of age, 70/sex/dose, for 6 hours/day, 5 days/week. Interim sacrifices were carried out on 10/sex/group after 6 and 12 months on this regimen. Terminal sacrifices of the surviving 50/sex/dose were carried out after 2 years.

Test atmospheres were generated by directing Telone fluid into "J-tube" assemblies, exposing it to preheated compressed air at 65°C, then sending the resultant ~~gas~~ vapor into the exposure chamber where it was mixed with air to generate the required concentration. Nominal doses were 0, 5, 20 and 60 ppm, which corresponded to 0, 4.6, 18.4 and 55.2 ppm when the 92.1% purity was taken into account. Spectrophotometric analyses conducted at least once/hour during the exposure periods, showed the chamber concentrations to be essentially equal to the nominal concentrations. Distribution of vaporized test article in the chamber was shown in a separate analysis to vary between 0 and 7.7% of the target 1,3-D concentration.

Rats were monitored for appearance and clinical signs after each exposure period. Dead or moribund animals were necropsied as soon as possible. Examinations for palpable masses were conducted at 6 and 12 months, then monthly thereafter. Body weights were determined weekly for the first 13 weeks and approximately monthly for the remainder of the study. Hematology and clinical chemistry were analyzed at 2 years only using blood samples from 20/sex/dose. Urinalyses were conducted on up to 20/sex/dose within a week of sacrifice. At 2 years, fasted rats were sacrificed, necropsied, organ weights (brain, heart, kidneys, liver and testes) measured, and representative tissues prepared for histopathology.

Mortality, clinical signs and palpable masses. There were neither deaths nor overt clinical signs attributable to Telone exposure during the 2-year period of this study. Survival at 2 years in males was, at ascending doses: 46%, 56%, 60% and 56%. Survival in females was 60%, 52%, 76% and 72%. There also was no effect on the appearance of palpable masses at any dose.

Body weights. A statistically significant suppression of mean body weight of about 5% was noted in 60-ppm males by test day 13, extending through day 453, and in 60-ppm females by test day 6, extending through day 327. Statistically significant reductions of ~3-4% were also noted at 20 ppm in males on days 117, 201, 229 and 327, though these were not nearly as consistently observed as at the high dose. 20-ppm females did not exhibit any statistically significant body weight reductions. Mean body weights for the first 34 days of the study are shown in detail in Table III.12, along with the weights at one year (day 369) and two years (day 733).

[2] Test article composition: 92.1% 1,3-dichloropropene (*cis*: 49.5%; *trans*: 42.6%); 0.7% 1,2-dichloropropane; 1.8% 1,3-dichloropropane; 1.1% 1-chlorohexane; ~4.3% mixed isomers of chlorohexene, chlorohexane and trichloropropene. ~2% epoxidized soybean oil acted as a stabilizer.

Hematology, clinical chemistry and urinanalysis. Hematologic parameters were unaffected by Telone exposure at any dose. Clinical chemistry parameters were unaffected in males. 60-ppm females showed slight, statistically significant reductions in total protein (g/dl at ascending doses: 6.3±0.5, 6.0±0.5, 6.1±0.4, 5.9±0.5*; *α=0.05) and in albumin (g/dl: 3.4±0.3, 3.3±0.3, 3.3±0.3, 3.2±0.2*). These effects were, in any case, not toxicologically significant. Urinalysis was also unaffected by Telone exposure.

Organ weights. With the exception of a slight, statistically significant decrease in female absolute brain weight at the high dose (at ascending doses, in grams: 1.869±0.044, 1.871±0.037, 1.875±0.048, 1.841±0.048*; *α=0.05; no effect on relative brain weight), there was no statistically significant change in any organ weight at any Telone concentration.

Gross pathology and histopathology. There were no gross pathological lesions that were unambiguously associated with Telone exposure. Histopathology revealed decreased thickness or erosions of the olfactory epithelium and fibrosis underlying the olfactory mucosa at 60 ppm in both sexes (Table III.13). One 20-ppm male exhibited both decreased thickness and erosion of the olfactory epithelium that was considered to be caused by Telone exposure. These lesions corresponded to anatomic levels 2-4, *i.e.*, from the incisive papilla (level 2) through the second palatal ridge (level 3) to the first upper molar teeth (level 4). There was no histopathology in the most anterior region, just posterior to the incisors (level 1).

A chronic NOEL of 5 ppm was determined based on the single 20-ppm male with nasal epithelial histopathology. It should be noted that body weight decreases at the high dose of 60 ppm were noted by day 6 in females and by day 13 in males, and thus were pertinent to the evaluation of potential risks arising from acute or short term exposures. For calculation of the appropriate Human Equivalent Concentrations, see section IV.F. below.

This study was considered to be acceptable according to FIFRA guidelines.

Table III. 12 Effect of daily Telone II inhalation exposure on body weights in Fischer 344 rats during the first month and at one and two years (Lomax *et al.*, 1987)

	Telone concentration, ppm							
	Males (grams)				Females (grams)			
Day	0	5	20	60	0	5	20	60
-1	150.0±14.9	151.8±14.3	153.3±14.3	148.8±16.7	108.3±4.1	107.9±4.0	107.6±4.5	107.4±5.3
6	179.4±12.3	180.3±12.1	182.7±11.2	172.4±17.5	126.1±4.7	125.5±4.4	126.6±4.9	121.0±5.7*
13	201.8±10.6	204.7±10.6	203.1±10.0	193.1±14.7*	135.4±4.6	135.1±4.8	135.6±5.5	129.0±5.3*
20	225.6±10.6	230.0±10.2	225.2±11.2	214.7±15.2*	147.1±5.3	147.8±5.4	146.7±6.6	139.1±5.7*
27	240.5±10.3	245.0±10.6	239.7±12.4	231.1±14.7*	155.6±6.6	155.4±5.6	154.8±7.1	147.9±6.7*
34	255.3±11.1	259.3±11.6	235.4±13.4	246.2±14.4*	162.3±7.4	163.5±6.6	163.8±7.6	155.5±6.6*
369	425.1±16.9	422.9±21.0	418.5±35.8	412.5±22.8[@]	242.2±17.2	246.9±19.7	245.8±15.3	240.8±14.2
733	413.5±44.1	396.5±42.1	391.2±40.6	389.8±39.8	282.5±27.3	286.9±31.3	281.0±26.5	288.9±36.0

* Statistically different from control mean, Dunnett's test, α=0.05

[@] Statistically different from control mean, Wilcoxon's test, α=0.05

Table III. 13 Non-neoplastic histopathology following 2 years of daily inhalation exposure to Telone in rats (Lomax *et al.*, 1987)

Parameter	Telone concentration (nominal [a]; ppm)							
	Males				Females			
	0	5	20	60	0	5	20	60
Nasal tissues								
Decreased thickness, olfactory epithelium	0/50	1/50	1/50	20/50*	0/50	0/50	0/50	15/49*
Erosion, olfactory epithelium	0/50	0/50	1/50	15/50*	0/50	0/50	0/50	6/49
Fibrosis, olfactory submucosa	0/50	0/50	0/50	6/50	0/50	0/50	0/50	2/50

*Different from control mean by Yate's chi-square pairwise test, alpha=0.05.

[a] Nominal concentrations corresponded to actual test article concentrations of 0, 4.6, 18.4 and 55.2 ppm.

2. Mouse inhalation

Stott (1987) exposed 70 B6C3F1 mice / sex/ dose to Telone II [3] vapors in 14 m^3 live-in chambers for 6 hr/day, 5 days/week. Test atmospheres were generated by directing Telone fluid into "J-tube" assemblies, exposing it to preheated compressed air at 65°C, then sending the resultant ~~gas~~vapor into the exposure chamber where it was mixed with air to generate the required concentration. Analytical concentrations were measured once/hr by spectrophotometry. Adequate chamber distribution was maintained within ±13%. Measurements at 9, 12, 18 and 23 months established the stability of the 1,3-D in the test article preparation.

Ten mice/sex /dose were sacrificed for necropsy at 6 and 12 months, with 50/sex/dose scheduled for the full 24-month exposure. The total number of exposure days was 510. The nominal doses were 0, 5, 20 and 60 ppm, which after correction for the purity of 92%, amounted to doses of 0, 4.6, 18.4 and 55.2 ppm. Mice were monitored for clinical signs at least once/day during the work week. Dead or moribund animals were necropsied as soon as possible. Body weights were determined weekly for the first 13 weeks and approximately monthly for the remainder of the study. Hematology and clinical chemistry were analyzed at 2 years only using blood samples from 20/sex/dose. About 50 tissues in each survivor were examined for gross pathology and histopathology at the end of 24 months.

Mortality and palpable masses. Telone exposure at the air concentrations used in this study had no clear effects on mortality relative to controls. Percent survival in males at 2 years was, at ascending doses: 90, 88, 90 and 94. Percent survival in females at 2 years was: 84, 88, 96 and 80. While males tended to have more palpable masses than females, no discernable effect of dosing was noted for this parameter.

Body weight. Body weight gains in males were statistically suppressed at the high dose during the first week (gains at ascending doses: 2.6, 2.5, 1.6 and 1.2* g; $p \leq 0.05$), but had resolved by the second week (1.4, 1.2, 1.6 and 1.6 g). Complete body weight data for the first month and at one and two years appear in Table III.14. Interestingly, statistically significant mean body weight deficits in males between controls and 60-ppm resurfaced by exposure day 202 and were maintained throughout the remainder of the study, such that at 2 years the mean weights at 60-ppm were 7% less than the controls. Females also sustained a slight, though statistically significant mean weight gain reduction during the first week at the mid and high doses (ascending doses: 2.0, 1.7, 1.5* and 0.7* g; $p \leq 0.05$). Like males, mean body weight in 60 ppm females was statistically suppressed from day 153 through the end of the study, when the weight deficit was 10%. The later body weight deficits were also of unclear toxicologic significance. Other studies (*eg.*, Gollapudi, 1998) suggest that decreases in food consumption at similar Telone concentrations may be responsible for such effects, though this parameter was not monitored in the current study.

[3] Telone II consisted of 92.1% 1,3-D (*i.e.*, 49.5% *cis* and 42.6% *trans* I,3-D, along with 0.7% 1,2-dichloropropane, 1.8%1,3-dichloropropane, 1.1% 1-chlorohexane and the remainder a mixture of isomers of chlorohexane, chlorohexene and dichloropropene). Epoxidized soybean oil (2%) acted as a stabilizer.

Hematology and serum chemistry. Hematology and serum chemistry analyses conducted at 2 years revealed several statistically significant changes in males, including lowered RBC numbers (9.31 *vs.* 8.79* x 10^6 / ml; $p \leq 0.05$), lowered hematocrit (37.5% *vs.* 35.8%*), increased serum urea nitrogen (22 *vs.* 26* mg/dl), increased alkaline phosphatase activity (48 *vs.* 53* mU/ml) and decreased serum globulin (2.6 *vs.* 2.3* g/dl). These changes were considered to be small and thus of uncertain toxicologic significance [4]. No such changes were observed in females.

Organ weights. Statistically significant reductions on the order of 10-15% in organ weights were noted at the high dose in heart (male absolute & relative to body weight, female absolute), kidney (male absolute & relative) and liver (male absolute), while increases were noted in brain (male relative, female absolute). Of these, only kidneys presented a histopathologic correlate (see below).

Gross pathology. The incidence of one or more lung masses / male rat increased in the 20-ppm and 60-ppm males relative to controls (incidence at ascending doses: 5/50, 3/50, 9/50, 14/50). Females did not show a similar profile. Lacrimal and uterine mass / nodules appeared elevated in all treatment groups, though dose responsiveness was not evident and toxicologic significance was unknown. Increased lymph node size that was attributed to lymphoreticular tumors was noted at the high dose in females (0/50, 1/50, 1/50, 3/50). However, histopathology did not show an increase in lymphoreticular tumors with dose in either sex. Pale liver and pale kidney were noted in 2/50 each among the high-dose females. Telone dependence was unclear. Low power microscopic examination of the urinary bladders showed roughened-irregular-opaque surface in 60-ppm males (incidence: 0/50, 1/50, 0/50, 6/50) and in 20- and 60-ppm females (3/50, 5/50, 20/50, 30/50).

Non-neoplastic histopathology. Non-neoplastic histopathology was noted most prominently in the urinary bladder, respiratory epithelium, olfactory epithelium and non-glandular stomach. In large measure, these signs reflected hyperplastic or hypertrophic responses to erosive or irritational effects of Telone (Table III.15). Decreased vacuolization was also noted at the high dose both in male kidneys (9/50, 8/50, 8/50, 29/50*) and in female livers (10/50, 9/50, 11/50, 24/50*). The investigators stated that "these changes were consistent with a relative decrease in cellular lipid and glycogen content of renal tubular epithelia and hepatocytes, respectively" (study p. 24), but did not feel that they impacted the health of the animals. Renal tubular mineralization also decreased in males at the mid and high doses (26/50, 35/50, 9/50*, 5/50*).

For urinary bladder, the following details were noted (quote from study pp. 21-22):

[4] The report seems to suggest that the increased serum UN content may have toxicologic significance: "….*with the possible exception of serum UN*, these small changes were not considered to reflect an adverse effect of treatment upon the health of the animals and probably represent normal variations in these measurements." (report, p. 19; emphasis added). However, it does not further comment. One could speculate that increased serum UN might represent a catabolic state, *i.e.*, one in which proteins are being broken down, though no further evidence for this is presented.

> "…urinary bladder effects consisted of an exposure-related increase in the occurrence and severity of hyperplasia of the transitional epithelium….of nearly all high exposure group male and female mice and in several male and female mice exposed to 20 ppm…. In most cases this lesion was characterized by a diffuse, smooth surfaced thickening of the epithelium (simple hyperplasia)…. Small focal areas of epithelia as much [as] 5-fold thicker than controls were also occasionally noted and, in several mice, the thickened epithelium contained downgrowths which reached into the submucosa but did not penetrate the basement membrane (nodular hyperplasia)… In general, more severe effects were noted in females than males… An inflammatory reaction was often associated with the hyperplastic response of the urinary bladder mucosa in female mice."

For nasal mucosa and respiratory epithelium (study pp. 22-23):

> "Nasal mucosal effects consisted of hypertrophy and hyperplasia of the respiratory epithelium and degeneration of the olfactory epithelium in nearly all high exposure group male and female mice. Hyperplasia of the respiratory epithelium was also noted in a majority of female mice exposure to 20 ppm Telone II soil fumigant. In all cases, both types of nasal lesions involved only approximately 10% or less of the respective epithelium affected and thus, both were considered to be of minimal severity… Respiratory epithelial lesions were characterized by their bilateral occurrence in a symmetrical pattern at the rostral aspects of the nasal cavity, primarily involving the mucosa of the nasal turbinate bone and the dorsal lateral aspect. Lesions of the olfactory epithelium also occurred bilaterally in a symmetrical pattern, primarily involving the mucosa of the dorsal portion of the nasal septum, dorsal wall of the nose and the ends of the endoturbinates which protrude into the dorsal meatus of the nasal cavity. Affected mucosa was thinned from its normal 6-8 cell thickness to as little as a single cell layer and the arrangement of the nuclei was disorganized and irregular in appearance…"

For the nonglandular stomach (study p. 23):

> "Treatment-related effects in the nonglandular portion of the stomach consisted of hyperplasia of the epithelium which was statistically identified in high exposure group male mice only. This lesion was characterized by a thickening of the stratified squamous mucosa, accentuation of the rete pegs and hyperkeratosis of the stratified squamous mucosa… Mononuclear inflammatory cells were often noted in the submucosa and small foci of granulocytic cells or ulcers were also occasionally seen in the thickened portions of the mucosa."

It seemed likely that the stomach histopathology described here resulted from movement of inspired Telone from the respiratory tract into the stomach.

Neoplastic histopathology. The incidence of presumptively benign bronchioloalveolar adenomas was elevated at the high dose in males (22/50 vs. 9/50 in controls; Table III.16). Treatment-

related elevation at the mid dose was also plausible (13/50), though this was not certain in view of the high control incidence, both within the study (18%) and in 7 previous chronic studies (7-32%). These tumors were described as follows (study p. 24):

> "Grossly, these tumors appeared as small (≤0.5 cm), pale, firm masses that were well demarcated from the surrounding lung tissue. Histologically, the tumors replaced the normal lung parenchyma and compressed the adjacent tissue. Tumor morphology did not vary with exposure level and did not appear to influence the survival of affected animals."

There was no evidence for treatment-related bronchioloalveolar tumors in females. Modeling of the dose-*vs*.-incidence rate of these tumors in males to establish the cancer potency appears below in section IV.

The incidence of lacrimal gland tumors also increased in all treated males (Table III.16), though the investigators discounted the possibility of a treatment relation. They based their opinion on the lack of statistical significance and dose responsiveness, as well as the laboratory's historical control range of 2-16%, which with the exception of the mid-dose rate of 20%, encompassed the values observed in the study. They felt that the apparent increase was an artifact of an "unusually low" control incidence rate. At any rate, since the lacrimal gland data lacked dose responsivity, they were not amenable to modeling. Further analysis was thus not carried out.

A chronic NOEL of 5 ppm was determined in this study based on the following observations at 20 ppm: body weight gain deficit during the first week in females and roughened-irregular-opaque urinary bladder surface in females (gross pathology), as well as hyperplasia / hypertrophy of the urinary bladder transitional epithelium and hyperplasia of the respiratory epithelium. Both histopathologic signs were observed in males and females. It should be noted that body weight decreases were noted by day 7, and thus were pertinent to the evaluation of risks arising from acute or short term exposures. Telone also induced bronchioloalveolar adenomas in males at 60 ppm, with a possible increase at 20 ppm (see discussion above). The co-occurrence of nasal respiratory epithelial hyperplasia / hypertrophy and bronchioloalveolar adenomas suggested that they were induced by 1,3-D as part of the same or similar process. For calculation of the appropriate Human Equivalent Concentrations, see section IV.F. below.

This study was acceptable according to FIFRA guidelines.

Table III. 14 Effect of daily Telone II inhalation exposure on body weights in B6C3F1 mice during the first month and at one and two years (Stott *et al.*, 1987)

	Telone concentration, ppm							
	Males (grams)				Females (grams)			
Day	0	5	20	60	0	5	20	60
-2	21.8±1.6	21.7±1.6	22.3±1.7	21.8±2.2	19.0±1.5	19.1±1.1	18.8±1.6	19.0±1.4
7	24.4±1.2	24.2±1.3	23.9±1.2	23.0±1.6*	21.0±1.1	20.8±1.0	20.3±1.2*	19.7±1.0*
13	25.8±1.4	25.4±1.5	25.5±1.2	24.6±1.8*	22.1±1.3	22.0±1.0	22.2±1.4	21.6±1.5
20	26.0±1.4	25.9±1.7	26.3±1.6	25.4±1.6	22.1±1.2	22.3±0.9	22.3±1.3	22.0±1.3
27	26.1±1.5	26.2±1.6	26.7±1.3	25.6±1.9	22.4±1.5	23.0±1.1	23.1±1.3*	22.8±1.4
34	26.7±1.6	26.4±1.7	27.7±1.9*	26.6±1.9	22.8±1.5	23.4±1.0	23.8±1.4*	23.5±1.4*
370	31.2±1.7	31.9±2.1	31.6±1.6	29.7±1.9*	28.1±1.7	28.3±1.6	28.1±1.8	27.3±1.4*
734	30.7±1.9	31.3±2.1	30.8±1.7	28.6±1.6*	29.6±2.7	30.0±2.7	29.1±2.2	26.8±2.9*

* Statistically different from control mean, Dunnett's test, α=0.05

Table III. 15 Non-neoplastic histopathology following 2 years of daily inhalation exposure to Telone in B6C3F1 mice (Stott *et al.*, 1987)

	Telone concentration (nominal [a]; ppm)							
	Males				Females			
Parameter	**0**	**5**	**20**	**60**	**0**	**5**	**20**	**60**
Urinary bladder								
Hyperplasia, simple, mucosa								
■ very slight	4/47	7/48	7/48	16/47*	1/47	3/46	13/48*	5/45
■ slight	0/47	0/48	3/48	18/47*	0/47	1/46	6/48*	18/45*
■ moderate	0/47	0/48	0/48	2/47	0/47	0/46	0/48	19/45*
Hyperplasia, nodular, mucosa								
■ slight	0/47	0/48	1/48	0/47	0/47	0/46	0/48	0/45
■ moderate	0/47	0/48	0/48	1/47	0/47	0/46	2/48	2/45
Hyperplasia, simple or nodular								
■ any severity	4/47	7/48	11/48	37/47*	1/47	4/46	21/48*	44/45*
Inflammation, chronic								
■ slight or moderate	0/47	0/48	0/48	0/47	0/47	1/46	2/48	7/45*
■moderate or severe	0/47	0/48	0/48	2/47	0/47	0/46	4/48	1/45
■ any severity	0/47	0/48	0/48	2/47	0/47	1/46	6/48*	8/45*
Nasal respiratory epithelium								
Hypertrophy & hyperplasia, nasal resp. mucosa, bilateral								
■ very slight	5/50	1/50	4/50	38/50*	4/50	4/50	28/50*	39/50*
■ slight	0/50	0/50	0/50	10/50*	0/50	0/50	0/50	10/50*
■ any severity	5/50	1/50	4/50	48/50*	4/50	4/50	28/50*	49/50*
Olfactory epithelium								
Degeneration bilateral								
■ very slight	1/50	0/50	1/50	32/50*	0/50	0/50	1/50	29/50*
■ slight	0/50	0/50	0/50	16/50*	0/50	0/50	0/50	16/50*
■ any severity	1/50	0/50	1/50	48/50*	0/50	0/50	1/50	45/50*
Non-glandular stomach								
Hyperplasia, often accompanied by chronic inflammation, focal or multifocal	0/50	3/50	1/50	8/50*	0/50	0/50	0/50	2/50

*Different from control mean by Yate's chi-square pairwise test, alpha=0.05.

[a] Nominal doses corresponded to analytically determined doses of 0, 4.6, 18.4 and 55.2 ppm.

Table III. 16 Neoplastic histopathology following 2 years of daily inhalation exposure to Telone in mice (Stott *et al.*, 1987)

	Telone concentration (nominal [a]; ppm)							
	Males				Females			
Parameter	**0**	**5**	**20**	60	**0**	**5**	**20**	**60**
Lung								
Bronchioloalveolar adenoma, benign								
■ one	9/49 [b]	6/50	11/49	20/50*	3/50	3/50	4/50	3/50
■ two	0/49	0/50	2/49	2/50	0/50	0/50	1/50	0/50
■ three	0/49	0/50	0/49	0/50	1/50	0/50	0/50	0/50
■ all	9/49	6/50	13/49	22/50*	4/50	3/50	5/50	3/50
Lacrimal gland								
Tumor, benign	1/49 [b]	6/50	10/49	5/50	6/50	3/50	3/50	3/50

*Different from control mean by Yate's chi-square pairwise test, alpha=0.05.

[a] Nominal concentrations corresponded to actual test article concentrations of 0, 4.6, 18.4 and 55.2 ppm.

[b] Tumor incidence is expressed per "at risk" animal. In this study, 2 animals (one control and one at 20 ppm, died without tumors during the first year and thus were not considered to be at risk. Consequently, those doses contained 49, not 50, animals.

3. Rat dietary

The following dietary administration study is included in this analysis because it provides independent support for oncogenic action of 1,3-D.

Stott (1995) administered Telone II [5] on a daily basis for up to 24 months to Fischer 344 rats---50/sex/dose---as a microencapsulated formulation mixed into feed. Doses were at 0, 2.5, 12.5 or 25 mg/kg/day (mean measured Telone consumption in males: 0, 2.5, 12.7, 25.4 mg/kg/day; females: 0, 2.5, 12.7, 24.8 mg/kg/day). An additional 10/sex/dose were sacrificed at 12 months. The following parameters were monitored: ophthalmology (before exposure and at 1 and 2 years); daily cageside observations; weekly clinical evaluations; body weights and weight gains (weekly for the first 13 weeks, approximately monthly thereafter); feed consumption (weekly for the first 13 weeks, then for a 1-week period per month thereafter); feed efficiency, hematology, clinical chemistry and urinalysis (at 6 and 12 months from the interim sacrifice group, then at 18 and 24 months from the main group); and upon sacrifice, organ weights, gross pathology, and histopathology.

Mortality, ophthalmology and in-life observations. Death rates were unaffected by dosing. All rats designated for interim sacrifice survived to the 1-year sacrifice point. By study termination, percent mortality at increasing doses was 26, 32, 32 and 32 in males and 22, 30, 38 and 22 in females. Neither ocular effects nor Telone-related in-life signs were noted.

Body weights. Body weights were statistically reduced at the high dose from the time of first measurement throughout the study in both sexes, reaching 9-16% reductions in males from 13 weeks forward and 10-15% in females from 73 weeks forward. Statistically significant reductions at the mid dose were noted starting at day 92 in males (3-9%) and in females (6-9%) at starting at day 649. A very slight reduction in mean body weight was evident in both sexes by study termination at the low dose, but statistical significance was not achieved at any point. By study termination on day 727, mean male body weights were (in grams) 384.2±39.9, 374.3±50.2, 364.0±35.6 & 335.1±38.5*, while female weights were 284.5±33.3, 280.5±32.5, 260.4±33.3* and 244.3±25.5* (*α=0.05).

Statistical significance of body weight gain reductions mirrored those of absolute body weights.

Feed consumption. Feed consumption was slightly reduced at the high dose in both sexes, though never achieving statistical significance.

Clinical pathology. Despite the occasional statistically significant detection in hematology, urinalysis and clinical chemistry, there was no indication of toxicologic significance. The only possible exceptions to this were statistically significant decreases in triglyceride levels at the mid

[5] The initial formulation was 96.0% 1,3-D (50.7% cis, 45.1% trans). It was added to microcapsules consisting of 80% starch / 20% sucrose. Loaded microcapsules contained 38.7% Telone II by weight. Stability was confirmed periodically during the study.

and high doses in males (12 and 24 months) and females (6, 18 and 24 months), an effect that was attributed by the investigators to decreased body weight.

Organ weights. Interim (1-yr) sacrifices in males showed the following statistically significant results at the high dose: reduced absolute heart and liver weights, and elevated relative (to body weight) brain, kidney and testes weights. Mid dose male relative brain weights were also elevated. Interim sacrifices in high dose females showed elevated relative brain, heart, kidney and liver weights. No statistically significant effects were noted in mid dose females.

Terminal (2-yr) sacrifices in males showed the following statistically significant results: reduced absolute heart and liver weights, and elevated relative brain and kidney weights. Mid dose male relative brain and kidney weights were also elevated. Terminal sacrifices in high dose females showed reduced absolute adrenal and heart weights, elevated absolute brain weights, and elevated relative brain, heart, kidney and liver weights. Mid dose females showed elevated relative brain, heart, kidney and liver weights.

As no histopathological correlates were identified to underlie these changes, they were considered by the investigators as secondary to the reduced body weights. The only exception was the elevated absolute brain weights in terminal high dose females, which were attributed to unusually low control brain weights.

Gross pathology interim (1-yr) sacrifice. No dose-dependent gross pathological lesions were noted among the 1-yr sacrifices.

Gross pathology terminal (2-yr) sacrifice. The following lesions were noted with a possible relationship to high-dose treatment in the 2-yr sacrifices: dark multifocal area of the liver in males (2/50, 2/50, 11/50, 7/50); roughened liver surface in females (3/50, 3/50, 8/50, 11/50); multifocal erosion and/or ulcers of the stomach glandular mucosa in males (6/50, 8/50, 8/50, 13/50). Several other lesions common to aging rats were noted, though without connection to dose.

Histopathology, interim (1-yr) sacrifice. About half of the mid dose and almost all of the high dose interim sacrifices exhibited basal cell hyperplasia of the non-glandular or squamous portions of the stomach, graded as very slight (♂: 0/10, 1/10, 5/10, 6/10; ♀: 0/10, 0/10, 2/10, 7/10), slight (♂: 0/10, 0/10, 2/10, 4/10; ♀: 0/10, 0/10, 1/10, 2/10) or any severity (♂: 0/10, 1/10, 7/10, 10/10; ♀: 0/10, 0/10, 3/10, 9/10),. This lesion is described by the investigators in the following way (study p. 38):

> "This effect was characterized by prominence of the basal or deepest layers of the mucosa due to increased cytoplasmic basophilia along with an apparent increased number of cell layers in the basilar portion of the mucosa. The normal round nuclei found in these basal cells were oval with the long axis perpendicular to the basement membrane. The basal cells were crowded together into a layer generally two or three cells thick rather

> [than] the single layer normally present. Additionally, there was a slight prominence of mononuclear cells at the basement membrane. These cells appeared to consist of endothelial, fibroblast and inflammatory cells."

No other toxicologically significant histopathological effects attributable to Telone were noted in the interim sacrifices.

Histopathology, non-neoplastic, terminal (2-yr) sacrifice. Non-neoplastic histopathology was noted in the kidney (increased tubular mineralization and pigmentation, high dose females), liver (increased foci of hepatocellular eosinophilic cells, possibly all doses, females) and stomach (non-glandular basal cell hyperplasia, top two doses, both sexes). The latter sign was similar to that described above for the interim sacrifices. Incidences of these lesions are noted in Table III.17.

Histopathology, neoplastic, terminal (2-yr) sacrifice. Benign liver adenomas were increased in both sexes at the high dose, though only achieving pairwise statistical significance in males. Incidence in females was statistically significant for trend. In addition, a non-statistically significant increase in benign liver adenomas was noted in mid dose males and was suggestive of a treatment effect. In addition, one high-dose male showed a malignant primary hepatocellular carcinoma. Finally, there was an apparent increase in benign uterine endometrial stromal polyps at the high dose, though statistical significance was not achieved.

In conclusion, the effects of Telone on body weight, liver histopathology (with a notable increase in benign adenomas at the high dose in males and females and possibly the mid dose in males) and stomach histopathology (non-glandular basal cell hyperplasia, top two doses, both sexes) were considered to be likely related to exposure. A definitive NOEL was not established, as the hepatocellular eonsinophilic foci may have been elevated in number and/or degree at all dose levels. The LOEL was, therefore, 2.5 mg/kg/day (the lowest dose tested). In addition, this study showed that Telone II induced benign liver adenomas in both sexes at 25 mg/kg/day and possibly in males at 12.25 mg/kg/day.

This study was considered to be acceptable by FIFRA guidelines.

Table III. 17. Histopathologic observations following 2 years of daily dietary exposure to Telone in Fischer 344 rats (Stott *et al.*, 1995)

Parameter	Telone dose (mg/kg)							
	Males				Females			
	0	2.5	12.5	25	0	2.5	12.5	25
Kidney								
Mineralization, tubules(s), multifocal, very slight	7/50	6/50	10/50	9/50	2/50	3/50	7/50	9/50*T
Increased pigment, tubule(s), any severity	6/50	6/50	7/50	9/50	4/50	1/50	6/50	13/50*T
Liver								
Foci of altered cells, eosinophilic, hepatocellular, very slight	29/50	25/50	18/50*	11/50*	12/50	24/50*	20/50	32/50*
Foci of altered cells, eosinophilic, hepatocellular, slight	3/50	11/50*	23/50*	24/50*	0/50	3/50	3/50	1/50
Foci of altered cells, eosinophilic, hepatocellular, moderate	0/50	0/50	2/50	1/50	0/50	0/50	0/50	0/50
Foci of altered cells, eosinophilic, hepatocellular, total	32/50	36/50	43/50	36/50	12/50	27/50*	23/50*	33/50*T
Adenoma, hepatocellular, benign, primary (1)	1/50	1/50	3/50	8/50*T	0/50	0/50	0/50	4/50
Adenoma, hepatocellular, benign, primary (2)	1/50	0/50	2/50	1/50	0/50	0/50	0/50	0/50
Adenoma, hepatocellular, benign, primary (3)	0/50	0/50	1/50	0/50	0/50	0/50	0/50	0/50
Adenoma, hepatocellular, benign, primary, total	2/50	1/50	6/50	9/50*T	0/50	0/50	0/50	4/50T
Carcinoma, hepatocellular, malignant, primary	0/50	0/50	0/50	1/50	0/50	0/50	0/50	0/50
Stomach								
Hyperplasia, basal cell, nonglandular mucosa, very slight	3/50	3/50	19/50*T	18/50*T	0/50	1/50	19/50*	33/50*T

Hyperplasia, basal cell, nonglandular mucosa, slight	0/50	0/50	1/50	12/50*	0/50	0/50	1/50	4/50
Hyperplasia, basal cell, nonglandular mucosa, total	3/50	3/50	20/50*	30/50*T	0/50	1/50	20/50*	37/50*T
Uterus								
Endometrial stromal polyp, benign, primary	n/a	n/a	n/a	n/a	12/50	13/50	10/50	24/50

*Statistically different from controls by Yate's chi-square pairwise test (α=0.10, two-sided; α=0.05, one-sided)

T: Linear trend by Cochran-Armitage test (α=0.02, two-sided; α=0.01, one-sided)

Table III. 18 Summary of NOEL and LOEL values from chronic inhalation studies on 1,3-D

Study type	NOEL	LOEL	Determining sign(s)	*Acceptability* Reference
■ Rat inhalation ■ 6 hr/day, 5 d/wk, 2 yr ■ 0, 5, 20 & 60 ppm	5 ppm	20 ppm	Nasal epithelial histopathology in one rat only @ 20 ppm	*Acceptable* (Lomax *et al.*, 1987)
■ Mouse inhalation ■ 6 hr/day, 5 d/wk, 2 yr ■ 0, 5, 20 & 60 ppm	5 ppm	20 ppm [a]	Roughened-irregular-opaque urinary bladder surface (♀), hyperplasia / hypertrophy of the urinary bladder transitional epithelium (♂ & ♀), hyperplasia of the nasal respiratory epithelium (♂ & ♀)	*Acceptable* (Stott *et al.*, 1987) [b]
■ Rat dietary ■ 0, 2.5, 12.5 & 25 mg/kg/day	Not determined	2.5 mg/kg/day (males)	Elevated incidence or severity of hepatocellular eonsinophilic foci	*Acceptable* (Stott *et al.*, 1995) [c]

[a] Critical LOEL value. BMCL modeling of the nasal respiratory histopathology incidence data resulted in a critical $BMCL_{10}$ of 6 ppm for this study (Appendix IV).

[b] This study was also used to calculate the oncogenic potency of 1,3-D based on the appearance of benign pulmonary adenomas.

[c] This study also showed 1,3-D dependent benign liver adenomas at 25 and possibly 12.5 mg/kg/day. Despite not utilizing the inhalation route, it is included in the table and in the assessment as a whole because it provides further evidence of the oncogenicity of 1,3-D.

E. GENOTOXICITY

Most of the genotoxicity studies mentioned below are summarized in Tables III.21 (registrant-submitted studies) and III.22 (studies conducted by the National Toxicology Program for the Gene Tox Program). The few that are not shown in these tables (Ghia *et al.*, 1993; Kitchin *et al.*, 1993; Kitchin and Brown, 1994; Kevekordes *et al.*, 1996) are discussed below in somewhat more detail than those already summarized in the tables. Also, supplemental discussion of genotoxicity issues is found in Appendices VI and VII.

Commercially available 1,3-D can contain autoxidation products that are direct-acting genotoxins. Even so, 1,3-D *cleared* of such impurities remains directly mutagenic (albeit less so) in the Ames Test using the base-substitution tester strain TA100. This is consistent with its direct reactivity with model nucleophiles, *e.g.*, 4-(4-nitrobenzyl)pyridine, and endogenous nucleophiles, *e.g.*, glutathione. 1,3-D induced gene mutations in mouse lymphoma cells at the TK locus but not in Chinese hamster ovary (CHO) cells at the HGPRT locus; since the positive effect in the former showed a preferential induction of small-colony-forming mutants, the gene mutations induced in that testing are assumed to result from large changes in genetic material such as chromosomal aberrations, as opposed to point mutations. Sister-chromatid exchanges, but not chromosomal aberrations, were induced in CHO cells exposed to 1,3-D both in the absence and presence of a metabolic activation system.

Regarding *in vivo* testing, 1,3-D induced sex-linked recessive lethals in *Drosophila* (fruit fly). Inhalation exposure of rats for 10 weeks did not increase the incidence of dominant-lethal mutations. Although *in vivo* testing for mutagenesis at the *lacI* transgene in lung and liver isolated from male Big Blue B6C3F1 mice exposed by inhalation to 1,3-D was negative, this testing is provisional due to the use of test conditions that presently are inadequate for concluding that the chemical is not mutagenic in this assay (discussed in Appendix VI).

Single intraperitoneal injection of 1,3-D into male B6C3F1 mice caused small, though reproducible and statistically significant, increases in chromosomal aberrations in bone-marrow cells and in the frequency of micronucleated polychromatic erythrocytes (PCE's) isolated from bone marrow. A greater effect was reported after single, oral dosing of female NMRI mice with either 187 mg/kg or 234 mg/kg 1,3-D. The mean incidence of bone-marrow micronucleated PCE's increased from 2.81 per 1000 PCE's in the corn-oil, pooled controls (a total of 6 females) to means of 15.3 and 14.9 per 1000 PCE's, respectively ($p < 0.01$ in both cases). By contrast, no micronucleus induction was seen with male NMRI mice dosed at 140 or 280 mg/kg (Kevekordes *et al.*, 1996). However, the increased response to oral dosing appears to be strain dependent. Single, oral dosing of CD-1 mice (both sexes) with 1,3-D in corn oil up to 380 mg/kg was negative for micronucleus induction in bone-marrow PCE's.

DNA damage was induced in two separate studies applying the alkaline-elution assay to cells isolated from organs of rats injected intraperitoneally or gavaged with 1,3-D. In Ghia *et al.* (Ghia

et al., 1993), three increasing doses of 1,3-D resulted in dose responses for DNA damage in liver and gastric mucosa; also, positive findings were seen in kidneys from the one dose level studied using different routes of exposure (gavage, i.p. injection). In liver, it was shown that pretreatment with an inhibitor of cytochrome P450 decreased the DNA damage induced by 1,3-D, indicating that some of the DNA damage depended on metabolic activation. In Kitchin *et al*. (Kitchin *et al.*, 1993) and Kitchin and Brown (Kitchin and Brown, 1994), significant DNA damage was induced in liver by 1,3-D given by gavage at 188 mg/kg and 564 mg/kg. However, unlike what was seen at the higher dose, the DNA damage at the lower dose was not accompanied by an increase in blood serum alanine aminotransferase activity or a decrease in P450 content. Therefore, the DNA damage at the lower dose appeared to result from direct genotoxicity and not through induction of hepatotoxicity.

Some of the metabolites of 1,3-D also are genotoxic. 1,3-D is epoxidated in the liver *in vivo* and by metabolic-activation systems *in vitro*. The resulting oxirane is a direct-acting mutagen to TA100; it hydrolyzes to give 3-chloro-2-hydroxypropanal, which may be the ultimate species that reacts with guanine in DNA. 1,3-D also hydrolyzes under aqueous conditions to 3-chloro-allyl alcohol (3-CAA). Although unreactive itself, 3-CAA can be metabolized by alcohol dehydrogenase to highly reactive 3-chloroacrolein (3-CA). Also, enzymatic hydroxylation of 1,3-D would produce 3-CA directly. While 3-CA is not mutagenic in the TA100 or TA98 strains, it induces frameshift mutations in *Salmonella* tester strain hisD3052. The differential mutagenicity with regard to tester strains may be an indication that 3-CA is a crosslinking agent. From structure-activity relationships for 3-substituted acroleins, reaction of 3-CA with DNA is expected to form fluorescent, unsaturated-ring adducts with guanine. 3-CA is also metabolized by aldehyde dehydrogenase to 3-chloroacrylic acid (3-CPA), which is mutagenic in TA97 and TA102 in the presence of an S9 activation system.

Collectively, these studies provide convincing evidence that 1,3-D, its oxidative metabolites and autoxidation products have genotoxic potential.

1. Dominant lethal study in rats

Thirty male Crl:CD®(SD) rats per treatment group were dosed by inhalation for 6 hr/day, 7 days/wk, 10 weeks duration, to Telone II at 0, 10, 60, and 150 ppm (Gollapudi *et al.*, 1997). The time-weighted mean analytical concentrations were 0, 10.1±0.70, 60.1±1.20 and 149.6±5.32 ppm. Dosing was based on a 4-week preliminary study that showed “excessive” weight loss at 200 ppm, and 150 ppm likely to be a maximum tolerated dose in a 10-wk study. Two lots of test article were used in the study, one containing 96.00% 1,3-D (49.3% *cis*, 46.7% *trans* isomers) and the other containing 95.6% 1,3-D (49.87% *cis*, 46.59% *trans* isomers). Negative pair-fed controls (matched to food consumption of high dose rats to control for reduced feed consumption at 150 ppm) and positive controls (single oral dose of cyclophosphamide given 48 hr prior to first mating period) were not housed in inhalation chambers. Each control group consisted of 30 males. There were two consecutive mating periods of 1 week each during weeks 11 and 12 (1

male / 2 females). On day 13 after the end of respective mating periods, females were euthanized. Corpora lutea were counted, and uteri were examined for numbers of live implants and resorption sites. Uteri of apparently non-pregnant females were stained with sodium sulfide and examined for early resorptions.

There were no clinical signs associated with treatment at any of the tested dose levels. Body weights were reduced in a dose-dependent and statistically significant manner at 60 and 150 ppm starting from the first measurement at day 8 (Table III.19). Weight gains at 60 ppm after day 8 were similar to controls (though absolute weights were often statistically reduced), while those at 150 ppm lagged to some degree. Paired feeding of untreated rats based on 150 ppm treated male consumption led to body weights comparable to those of 60 ppm males maintained in inhalation chambers [6]. Higher body weights in the cyclophosphamide rats (which were not dosed until 48 hr before mating) may represent normal variation between subpopulations or alternatively may reflect a difference between housing environments (possible effects of housing of 1,3-dichloropropene rats and standard controls in inhalation chambers compared to normal caging). Food consumption was reduced at 150 ppm at most time intervals. At 60 ppm, food consumption was reduced during the first week only (Table III.20). The body weight deficits at both doses may have been secondary to the deficits in food consumption or were the result of generalized stress. There was no increase in resorptions, consequently no evidence for a dominant lethal genotoxic effect.

The NOEL for short term change was 10 ppm based on decreased food consumption and decreased body weight at 60 and 150 ppm, particularly during the first week of treatment. It should be noted that the body weight decreases were noted after 7 days of exposure and thus may be pertinent to the evaluation of potential risks arising from acute or short term exposures. For calculation of the appropriate Human Equivalent Concentrations, see section IV.F. below. This study was deemed acceptable according to FIFRA guidelines.

[6] Initially the pair-fed rats were heavier than rats in other groups, reflecting the fact that these rats had one additional week of growth than the other rats in each column, due to study design.

Table III. 19 Body weights of male rats exposed to 1,3-D by inhalation for 10 contiguous weeks, 6 hr/day, 7 days/wk (Gollapudi *et al.*, 1997)

Days on Test	Treatment					
	1,3-Dichloropropene (ppm)				Cyclophosphamide	Pair-fed
	0	10	60	150	75 mg/kg/day	Controls [a]
-4	343.4±16.1	342.2±15.8	344.0±14.2	347.3±15.6^{+}	346.5±11.2	342.8±13.1
1	365.6±18.6	362.6±17.8	365.3±35.9	364.7±16.2^{+}	365.7±13.7	363.8±16.5*
8	374.5±20.7	372.5±20.4	359.2±20.3*	341.0±16.8*$^{+}$	388.0±17.6*	385.8±36.1*
15	393.1±24.4	388.7±22.7	380.1±20.7	347.3±19.4*$^{+}$	406.8±35.4*	394.5±19.6*
22	410.7±26.8	406.6±35.0	395.0±24.3*	354.4±20.9*$^{+}$	435.5±35.5*	396.1±18.8
29	430.1±29.5	427.5±28.3	412.8±35.4*	364.2±22.9*$^{+}$	454.3±28.4*	392.7±17.7*
36	444.8±32.7	443.3±31.2	426.4±26.2*	369.2±25.5*$^{+}$	482.2±31.3*	407.4±17.8$^{\$}$
43	461.3±34.6	457.4±32.0	437.3±26.7*	376.2±28.7*$^{+}$	491.4±34.6*	424.5±18.0$^{\$}$
50	469.4±36.6	466.6±33.7	448.3±28.5*	381.5±30.4*$^{+}$	500.6±36.2*	430.8±16.7$^{\$}$
57	479.3±38.2	481.6±35.6	458.7±30.2	385.6±30.7*$^{+}$	509.3±38.5*	445.8±17.1$^{\$}$
64	490.3±39.9	490.8±34.7	471.1±31.7	392.2±33.2*$^{+}$	522.1±40.0*	454.5±17.6$^{\$}$
70	501.6±40.7	499.4±37.7	474.8±31.6*	389.9±34.2*$^{+}$	530.2±41.6*	460.2±18.4$^{\$}$
79	511.7±37.8	509.2±35.4	493.2±31.7	435.3±32.4*$^{+}$	497.4±35.1	463.9±17.6$^{\$}$
85	520.3±37.7	539.9±34.6	500.9±32.1	456.1±32.1*$^{+}$	513.0±37.1	497.7±28.2
95	n/a	n/a	n/a	n/a	n/a	511.7±31.4

* Significantly different from the control mean, Dunnett's test, α= 0.05

$^{+}$ Significantly different from paired mean, α=0.05

$^{\$}$ Significantly different from the control mean, Wilcoxon's test, α=0.05

[a] Pair-fed controls were maintained in separate housing and started on test one week later than the parallel treated groups (see text footnote 6).

Table III. 20 Food consumption in male rats exposed to 1,3-D by inhalation for 10 contiguous weeks, 6 hr/day, 7 days/wk (Gollapudi *et al.*, 1997)

Days assessed	Treatment				
	1,3-Dichloropropene (ppm)				Cyclophosphamide
	0	10	60	150	75 mg/kg/day
1-8	25.7±1.6†	25.6±1.9	21.2±1.6	17.9±1.9	25.9±1.8
8-15	24.0±1.6	24.2±1.9	23.8±1.9	20.3±1.6	26.5±2.1
15-22	23.8±1.7	24.2±2.0	23.3±2.3	20.6±1.1	26.93.0
22-29	24.8±1.9	25.8±2.4	24.7±1.8	23.0±1.9	27.5±2.4
29-36	24.8±2.1	25.5±1.8	24.8±1.7	21.8±1.7	27.6±2.3
36-43	25.1±1.8	26.3±2.1	25.3±1.8	23.2±2.3	27.9±2.7
43-50	24.9±2.0	26.1±1.9	25.4±1.7	22.5±2.2	27.5±2.5
50-57	24.4±1.8	27.0±2.4	25.6±1.7	23.5±2.2	27.5±2.7
57-64	24.6±2.0	26.3±2.1	25.9±1.6	22.6±2.0	28.2±2.3
64-70	24.9±1.6	26.6±2.9	25.6±2.2	23.2±2.2	26.8±2.8

† Units of food consumption were not provided in the source table (p. 31). Values are most consistent with units of mg/kg/day, considering the approximation that rats consume about 1/20 of body weight per day after the period of rapid growth.

Note: The investigators did not apply statistical tests to the above data. However they noted that there was a clear high dose reduction and a transitory mid-dose reduction in food consumption. The DPR reviewer confirmed that the value for 60 ppm males over days 1-8 was highly significant by t-test, hence supportive of "subacute" LOEL.

2. Genetic damage in "Big Blue" mice

Gollapudi and Cieszlak (1997) dosed male Big Blue B6C3F1 mice, 5/group, by inhalation at 0, 10, 60, or 150 ppm of Telone II Soil Fumigant (96%), for 6 hr/day, 5 days/wk for 2 weeks. After an additional 17-day expression period, the mice were killed. Each mouse cell contained ~40 copies of a shuttle vector carrying the *lacI* gene, the *lacI* promotor, the *lacI* operator and the *αlacIZ* reporter gene. Mouse tissues (lung and liver) were then homogenized and the DNA collected, digested and packaged into phage particles using a proprietary system. The packaged DNA was added to plates containing *E. coli* host bacteria. Following incubation, the investigators counted the numbers of blue plaques compared to the total number of plaques to generate an index of mutations in the *lacI* gene [7]. Only controls and 150 ppm mice were evaluated. Results showed no increases in mutations in lung or liver. Functional positive control

[7] Blue plaques occur when a defective repressor protein allows transcription of the reporter gene, the product of which cleaves a chromogenic substrate (X-gal) in the medium.

tissues evidently derived from a single mouse which was treated with five daily doses of 15 mg/kg/day diethylnitrosamine in water 54 weeks before sacrifice.

This study was not considered to be acceptable by FIFRA standards, but was upgradeable pending clarifications regarding how the positive controls were run. Further discussion of this study appears in Appendix VI.

Table III. 21 Genotoxicity of 1,3-D in in vitro and in vivo assays – Registrant-submitted assays

Assay type	Dose or concentration	S9, +/ -	Result	*Acceptability* Reference
Mutagenicity, *in vitro*, bacterial assays				
***E. coli* strain B/r, Wp 2, Try / reverse mutation**	5000, 2500, 1000, 500, 250, 100, 25, or 0 µg/plate	±	Negative	*Unacceptable* [a] (Nomura Sogo Research Institute, 1978)
***Salmonella* plate assay (5 strains)**	0-5000 µg/plate [b]	±	Positive in several strains; negative in host-mediated assay	*Unacceptable* [c] (Nomura Sogo Research Institute, 1978)
***Salmonella* plate assay (TA 100 only)**	0-300 µg/plate [d]	±	Negative	*Supplemental* (Lawlor, 1996)
Mutagenicity, *in vitro*, mammalian cell assays				
CHO/HGPRT assay	250, 200, 150, 100, 50, or 0 mM - S9 (3 trials) 200, 150, 125, 100, 50, or 0 mM + S9 (1 trial)	±	Negative	*Acceptable* (Dow Chemical Company, 1986)
DNA damage, *in vitro*				
Unscheduled DNA synthesis in rat hepatocytes	$1x10^{-7}$ to $3x10^{-3}$ M concentration (solubility limit)	-	Negative	*Acceptable* (Dow Chemical Company, 1985a)
Adduct formation when ^{14}C-labeled test material was incubated with calf thymus DNA	Concentration not stated in one-liner	±	Negative	*Supplemental* (Stott *et al.*, 1997)
Cytogenetic or unspecified genetic damage, *in vivo*				
Inhalation genotoxicity in male "Big Blue" B6C3F1 mice	0, 10, 60 and 150 ppm, 5 days/wk, 6 hr/day	n/a	Negative	*Unacceptable* [e] (Gollapudi and Cieszlak, 1997)
CD1 mouse bone marrow micronucleus test	380, 115, 38, or 0 mg/kg by oral gavage, 5/sex/group, 24 or 48 hour sacrifice	n/a	Negative	*Acceptable* (Dow Chemical Company, 1985b)
Inhalation dominant lethal mutations in CD rats	30 male CD rats / group, inhalation 6 hr/day, 7 days/wk, 10 weeks @ 0, 10, 60, and 150 ppm	n/a	Negative	*Acceptable* (Gollapudi *et al.*, 1997)

[a] Design deficiencies

[b] Test article contained epichlorohydrin

[c] Single plates, epichlorohydrin possibly present

[d] Non-cytotoxic range

[e] Concern about positive control

Table III. 22 Genotoxicity of 1,3-D in assays conducted by National Toxicology Program

Assay type	Dose or concentration	S9 +/ -	Results	Reference
Mutagenicity, *Salmonella* tester strains				
Ames test using 20 min preincubation (TA100, TA1535, TA1537, TA98) and Aroclor-induced liver S9 from male rats *versus* male hamsters	0 (ethanol), 3→3333 µg/plate (0.027→30 µmole/plate)	+/ -	at 3 µmole w/o S9, revertants/plate ↑ by factors of 15 (TA100) and 13 (TA1535), much less ↑ when either S9 used; with TA98 w/o S9, weak response at 1 µmole	(Haworth *et al.*, 1983)
Mutagenicity, *in vitro*, mammalian cell assays				
L5178Y thymidine kinase$^{+/-}$ -3.7.2C mouse lymphoma cell mutation assay, without the use of metabolic activation (S9)	0 (ethanol), 27→216 µM (1st trial) and 0, 27→162 µM (2nd trial), 4 hr exposure 162 µM was toxic in 2nd trial	-	average mutant frequency ↑ by factors up to 12 and 8 in the two trials, with preferential induction of small colonies	(Myhr *et al.*, 1991)
Chromosomal damage, *in vitro*, Chinese hamster cells				
Sister-chromatid exchanges (SCE)	0 (DMSO), 9→269 µM (1st trial) and 0, 270→451 µM (2nd trial), 26 hr exposure, no S9 0 (DMSO), 27→269 µM (3rd trial), 2 hr exposure, with S9	+/ -	SCE's per cell ↑ by factors of 1.7, 2.6 and 1.7 at the highest concentrations in the three trials, respectively	(Loveday *et al.*, 1989)
Chromosomal aberrations	0 (DMSO), 44→442 µM (1st trial) and 0, 451→901 µM (2nd trial), 8 hr exposure, no S9 0 (DMSO), 90→451 µM (3rd trial), 2 hr exposure, with S9	+/ -	16% of cells with aberrations in the 1st trial at 442 µM, but not repeated in the 2nd trial; no ↑ in aberrations w/ S9; chromatid gaps in high % of cells w/o & w/ S9	(Loveday *et al.*, 1989)
Mutagenicity, *Drosophila melanogaster*				
Induction of sex-linked recessive lethals (SLRL) and reciprocal translocations (RT)	Males were fed for 72 hr on 5% sucrose (in 10% aqueous ethanol) containing 5570 ppm 1,3-D (freshly made each 24 hr), resulting in 33% mortality and 10% sterility	n/a	0.30% SLRL *vs.* 0.12% in controls ($p \leq 0.01$); 1 RT recovered in 6955 tests *vs.* $\geq$ 2 RT's needed to be considered significant	(Valencia *et al.*, 1985)

Chromosomal damage, *in vivo*, male B6C3F1 mice				
bone-marrow chromosomal aberration test, using a single IP injection, sacrifice 17 hr later	0 (corn oil), 50→200 mg/kg (1st trial) and 0, 200 mg/kg (2nd trial); 8 animals/dose for each trial	n/a	% cells w/ aberrations ↑ by factors of 2.8-2.9 ($p<0.01$) at 200 mg/kg each trial	unpublished data obtained from NTP Archives (K. Witt, 2015)
bone-marrow micronclеus assay, using a single IP injection, sacrifice 48 hr later	0 (corn oil), 100, 200 mg/kg (1st trial) and 0, 150, 250 mg/kg (2nd trial); 5-7 animals/dose each trial; 2000 polychromatic erythrocytes (PCE) scored per animal	n/a	‰ micronclеated PCE ↑ by factors of 2.3 and 3.6 ($p<0.01$) at highest doses in each trial; no increase if sacrificed at 24 hr or if 3 IP injections given	(Shelby *et al.*, 1993)

F. REPRODUCTIVE TOXICITY

1. Rat

Breslin (1987)exposed 30 rats/sex/group to Telone II (1,3-D: 91.2%) by inhalation to 0, 10, 30 or 90 ppm. The exposure regimen for F0 animals was 6 hours/day, 5 days/week for 10 weeks prior to breeding and 7 days/week, during breeding, gestation and lactation. F_1 pups selected to breed to produce F_2 litters received 12 weeks of pre-mating inhalation exposure, then exposures of 7 days/week during breeding, gestation and lactation. Exposure continued until sacrifice. This treatment schedule was continuous except that gravid females were not dosed from presumed gestation day 20 until lactation day 4. There were two generations with two littering periods each. Pups were not directly exposed, but were separated from dams during the 6-hour maternal exposures.

No adverse effects were observed on reproductive parameters, which are summarized in Table III.23. This resulted in a reproductive NOEL of $\geq$ 90 ppm. The parental NOEL was 30 ppm, based on decreased adult body weights (often statistically significant in males and occasionally significant in females) and nasal histopathology (slight hyperplasia of the respiratory epithelium in the majority of adults, and degeneration of the olfactory epithelium in many adults) at 90 ppm.

The study was considered to be acceptable according to FIFRA guidelines.

Table III. 23 Effect of inhalation exposure to 1,3-D on reproductive parameters in Fischer 344 rats (Breslin *et al.*, 1987)

F1a generation:

Reproductive parameter	1,3-D concentration (ppm) 0	10	30	90
# F0 females on study	30	30	30	30
# F0 females mated	30	29	27	29
# F0 males on study	30	30	30	30
# F0 males mated	28	23	24	25
Gestation index	22/22	22/22	22/22	26/26
Gestation survival index (%)	99.1	99.5	100	99.7
Mean live litter size (day 1)	10.3	9.8	10.7	11.4
Mean pup weight (day 1) (grams)	5.5	5.4	5.5	5.3
Mean live litter size (day 21)	7.9	7.3	7.9	7.7
Mean pup weight (day 21)	25.7	25.6	25.3	25.5

F1b generation:

Reproductive parameter	1,3-D concentration (ppm) 0	10	30	90
# F0 females on study	30	30	30	30
# F0 females mated	29	30	27	29
# F0 males on study	30	30	30	30
# F0 males mated	24	27	23	26
Gestation index	24/25	24/24	23/23	25/25
Gestation survival index (%)	98.1	99.6	99.3	100
Mean live litter size (day 1)	10.4	10.0	11.7	10.9
Mean pup weight (day 1) (grams)	5.2	5.1	5.1	5.2
Mean live litter size (day 21)	7.3	6.9	8.0	7.5
Mean pup weight (day 21)	28.0	27.2	27.7	27.1

F2a generation (F2b, data similar, not shown here):

Reproductive parameter	1,3-D concentration (ppm) 0	10	30	90
# F0 females on study	30	30	30	30
# F0 females mated	28	29	27	29
# F0 males on study	30	30	30	30
# F0 males mated	23	24	20	27
Gestation index	24/24	23/23	19/19	27/27
Gestation survival index (%)	100	98.8	100	99.6
Mean live litter size (day 1)	8.9	10.7	11.4	10.3
Mean pup weight (day 1) (grams)	5.3	5.1	5.0	5.0
Mean live litter size (day 21)	6.7	7.7	7.6	7.6
Mean pup weight (day 21)	24.4	24.6	24.8	24.3

G. DEVELOPMENTAL TOXICITY

1. Rat preliminary

Kloes (1983)conducted a preliminary whole-body inhalation study in pregnant Fischer 344 rats and New Zealand White rabbits with a purpose of determining the maximum tolerated dose of Telone II [8] for developmental toxicity in both species. This summary concerns only the rat phase of the study. Dose groups consisting of 7 or 8 pregnant rats were exposed to this test article at target concentrations of 0, 50, 150 or 300 ppm for 6 hr/day on gestation days 6-15. These corresponded to analytically determined concentrations of 0, 50±1, 148±8 and 301±3 ppm. Telone atmospheres were created by metering known amounts of liquid Telone into glass tubes followed by removal and vaporization with compressed air heated to 100°C, then dilution and mixing before introduction into the exposure chambers. Observations for toxic signs and mortality were conducted daily. Body weights were determined on gestation days (gd) 6, 8, 10 and 16. Food and water consumption were measured every 3 days starting on gd 6. Rats were

[8] 47.7% cis and 42.2% trans 1,3-D with the following impurities: 1.8% epichlorohydrin, 0.2% 2,3-dichloropropene, 1.4% 1,2-dichloropropane, 1.5% 1,3-dichloropropane and 2.2% 1-chlorohexane.

sacrificed on gd 16. Gross pathologic changes were noted immediately upon sacrifice. Necropsies included determinations of liver and kidney weights, number of implantations and resorbed fetuses, and early resorptions. Histopathologic analysis was not conducted.

Maternal effects

Mortality and clinical signs. One 300-ppm dam was found dead on gd 14, an effect likely due to Telone exposure. Clinical signs at 300 ppm included urine and fecal staining, nasal exudate and red crusty material around the eyes. Neither deaths nor clinical signs were noted for the 50 or 150 ppm groups.

Maternal body weights. Statistically significant body weight losses were recorded at all doses between gd 6 and 8. Weight loss continued at 150 ppm through gd 10, and at 300 ppm through sacrifice at gd 16. Statistically significant decrements in body weight gain were present at all doses for the entire gd 6-16 period (Table III.24). Total body weights were statistically reduced at 150 and 300 ppm on gd 8, 10 and 16.

Kidney and liver weights. While no differences were noted in absolute kidney weights, relative weights were statistically increased. This change was considered secondary to the effect on body weight. Liver weights appeared to decrease compared to controls at all doses, but since statistical significance was achieved only at 150 ppm, a Telone-based etiology was unclear. Relative liver weights were statistically elevated at the high dose, also secondary to the effect on body weight.

Food and water consumption. Food consumption was significantly less than controls for the gd 6-9 and 9-12 periods at 150 ppm, and for the gd 6-9, 9-12 and 12-15 periods at 300 ppm (Table III.25). It was not reduced at 50 ppm, which shows that the weight losses sustained over the first 3 days of gestation were not necessarily due to reduced consumption. Water consumption was significantly less than controls for the gd 6-9 period at 150 ppm and for the gd 6-9 and 9-12 periods at 300 ppm.

Gross pathology. Gross pathologic lesions were noted at 300 ppm only, with the possible exception of decreased thymus size in one animal at 150 ppm. These included stomach erosions or ulcers, decreased thymus size, strangulated or necrotic abdominal fat, decreased fat, alopecia and perineal soiling. In addition, most of these signs appeared in the decedent rat at 300 ppm. Incidence rates for the survivors appear in Table III.26. These effects were considered by the investigators to be stress related.

Embryo and fetal effects

Statistically significant increases in the percent implantations resorbed and the percent litters with resorptions, accompanied by a statistically significant reduction in fetuses/litter were noted at 300 ppm (Table III.27). Percent implantations resorbed may also have risen at 150 ppm, though the lack of statistical significance makes it difficult to clearly attribute to Telone exposure

at this dose. Statistically significant increases in percent preimplantation loss at 50 and 150 ppm were not considered Telone-related since implantation occurred before the onset of exposure on gd 6.

A maternal NOEL was not identified in this study since statistically significant body weight loss was observed at all doses on gd 6-8 and 6-16. A developmental NOEL was established at 150 ppm based on increased resorptions at 300 ppm. This NOEL was considered preliminary because of the low statistical power of this study and because, as noted in the previous paragraph, the percent implantations resorbed rose at 150 ppm, though without statistical significance. It should be noted that this study utilized repeated doses to establish these values. Nonetheless, the body weight decreases were noted after 3 days of exposure and thus may be pertinent to the evaluation of potential risks arising from acute or short term exposures.

This study was considered to be supplemental.

Table III. 24 Body weights and organ weights in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

	Telone (ppm)			
	0	**50**	**150**	**300**
	Maternal body weight (grams)			
Gestation day:				
■ 6	192.4±9.54	194.3±7.58	195.7±8.37	196.1±5.42
■ 8	195.8±9.03	189.9±8.19	181.4±7.46*	174.7±2.39*
■ 10	199.6±9.78	192.3±8.38	180.8±9.06*	156.8±3.78*
■ 16	218.1±7.79	207.6±11.57	186.9±6.41*	145.0±17.05*
	Maternal body weight gain (grams)			
Gestation days:				
■ 6-8	3.5±3.96	-4.5±2.51*	-14.3±3.74*	-21.4±5.04*
■ 8-10	3.8±2.49	2.4±3.10	-0.6±3.34	-18.0±2.41*
■ 10-16	18.5±4.39	15.3±4.13	6.1±11.80	-11.7±17.06*
■ 6-16 (Total)	15.8±3.53	13.3±7.18*	-8.8±13.38	-51.1±12.70*
	Maternal liver weight, gestation day 16			
■ **Absolute (grams)**	8.59±0.33	8.20±0.56	7.34±1.07*	7.17±1.37
■ **Relative** [a]	3.99±0.09	4.04±0.13	4.02±0.22	4.95±0.63*
	Maternal kidney weight, gestation day 16			
■ **Absolute (grams)**	1.43±0.08	1.51±0.08	1.47±0.07	1.48±0.10
■ **Relative** [a]	0.66±0.03	0.74±0.03*	0.81±0.05*	1.04±0.14*

* Dunnett's test, α=0.05

[a] Grams organ weight / 100 grams body weight

Table III. 25 Food and water consumption in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

	Telone (ppm)			
	0	**50**	**150**	**300**
	Food consumption (grams/day)			
Gestation days:				
■ 6-9	13±2	13±1	9±4*	3±3*
■ 9-12	14±1	14±1	10±3*	2±1*
■ 12-15	15±2	15±1	13±2	8±4*
	Water consumption (grams/day)			
Gestation days:				
■ 6-9	23±6	19±3	15±3*	10±4*
■ 9-12	28±3	26±2	27±4	11±14*
■ 12-15	24±2	29±4	31±7	24±7

* Dunnett's test, α=0.05

Table III. 26 Gross pathologic observations in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

	Telone (ppm)			
	0	**50**	**150**	**300**
Stomach				
■ Erosion, glandular mucosa	0/8	0/7	0/8	1/7
■ Ulcer, nonglandular mucosa, multifocal	0/8	0/7	0/8	2/7
Thymus				
■ Decreased size	0/8	0/7	1/8	7/7
Abdominal cavity				
■ Strangulated or necrotic fat	0/8	0/7	0/8	1/7
External and skin				
■ Alopecia, back	0/8	0/7	0/8	1/7
■ Perineal soiling	0/8	0/7	0/8	4/7

Table III. 27 Embryo and fetal parameters in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

Parameter	Telone (ppm) 0	50	150	300
# Bred	8	7	8	8
# Maternal deaths	0	0	0	1
% Pregnant	100 (8/8)	100 (7/7)	75 (8/8) [a]	100 (8/8)
# Litters	8	7	6 [a]	7
Corpora lutea / dam	11±1	11±1	12±2	11±2
Implantation sites / dam	11±2	10±2	9±3	11±2
% Preimplantation loss	3±7	19±19*	20±18*	6±14
Fetuses / litter	11±2	9±3	8±4	3±4*
Resorptions / litter	0.4±0.5	0.3±0.5	2±3	8±4
% Implantations resorbed	3 (3/87)	8 (5/65)	19 (11/57)	77 (61/79)*
% Litters with resorptions	38 (3/8)	43 (3/7)	50 (3/6)	100 (7/7)*
# Litters totally resorbed	0	0	1	3
Resorptions / litters with resorptions	1 (3/3)	1.7 (5/3)	3.7 (11/3)	8.7 (61/7)

*Different from control, α=0.05

[a] Two pregnancies at 150 ppm were detected only after uterine staining with sodium sulfide. Those animals did not produce litters.

2. Rat, complete

John (1983) exposed 30 mated female Fischer 344 rats to Telone II (90.1% 1,3-D, consisting of 47.7% trans and 42.4% cis isomers) by the inhalation route for 6 hr/day on gestation days (gd) 6-15. The study was conducted to identify possible effects on developmental processes during the period of major organogenesis in the rat. After co-housing one virgin female with one male, day 0 of pregnancy was marked by the appearance of sperm in a vaginal smear. Inhalation exposures occurred in whole-body chambers at target concentrations of 0, 20, 60 or 120 ppm, which were created by directing metered quantities of Telone into vaporization tubes, followed by removal of the tube contents into the chamber with a preheated compressed air stream. Mean analytical concentrations, determined once/hr by spectrophotometry, were essentially equivalent to the target concentrations. Standard observations in the dams were made for mortality / moribundity, body weights (gestation days 6, 9, 12, 16 and 21), food and water consumption (3-day intervals starting on gd 6), maternal liver and kidney weights (time of sacrifice). Rats were euthanized on gd 21. Pregnancy observations included number and, where applicable, position of fetuses *in utero*, live and dead fetuses, implantation sites, resorption sites and corpora lutea. Fetuses were examined for gender, weight, crown-rump length, gross external pathology, soft tissue alterations, skeletal alterations and head pathology.

Maternal effects

Mortality / moribundity. Neither deaths nor clinical signs were evident in the dams.

Body weight. There were dose-dependent, statistically significant body weight losses or weight gain decrements at all treatment levels that were manifest by gd 9 (the first measurement after starting the exposure regimen), even at the low dose (Table III.28). At the high dose, this amounted to a 5.4% weight loss during the first three exposure days. At the low dose, the animals gained only 1±4 grams, which was significantly reduced from the control gain of 4±4 grams (Dunnett's test, α=0.05). While the low dose animals began to gain weight at, or close to, control rates by gestation days 9-11, and by gestation days 12-15 at the mid dose, statistically significant reductions in maternal body weight were still evident at all doses at the end of gestation. It is likely that these body weight effects resulted from decrements in food consumption (next paragraph).

Food and water consumption. Food consumption was reduced in a dose-dependent, statistically significant manner at all doses through gd 17 (Table III.29). The reason for these consumption decrements was not clear. As Telone was not in the food, it is unlikely that palatability was the issue. It was plausible that the animals felt sickened after exposure to Telone and thus avoided food.

Water consumption was reduced only at the high dose, and only between gd 6 and 11.

Organ weights. Absolute liver weights were statistically reduced in all treatment groups, an effect that was probably secondary to the effect on body weight as suggested by the lack of effect on relative liver weight. While absolute kidney weights were not affected, relative kidney weight at the high dose was statistically elevated, also likely secondary to the body weight effect.

Embryo or fetal effects

Neither embryo nor fetal toxicity was noted upon caesarian section at any dose (Table III.30). Incidences of external, soft tissue, skeletal and skull bone alterations were also not affected by dosing, with the possible exception of delayed ossification of the vertebral centrum, a relatively common finding. There appeared to be a rise in this parameter at the high dose, though statistical significance was not achieved. This rise was likely secondary to the body weight effect.

The NOEL for maternal effects was <20 ppm based on the dose-dependent body weight and food consumption decrements noted at this dose. It should be noted that body weight decreases were noted by gd 8, and thus may be pertinent to the evaluation of potential risks arising from acute or short term exposures. The NOEL for developmental effects was 60 ppm based on the delayed ossification of the vertebral centrum noted in fetuses at 120 ppm. For calculation of the appropriate Human Equivalent Concentrations, see section IV.F. below.

This study was acceptable according to FIFRA guidelines.

Table III. 28 Body weights and organ weights in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (John *et al.*, 1983)

	Telone (ppm)			
	0	**20**	**60**	**120**
	Maternal body weight (grams)			
Gestation day:				
■ 6	185±7	184±9	187±8	186±8
■ 9	189±7	185±10	184±7	176±8*
■ 12	199±7	193±9*	190±8*	178±9*
■ 16	212±7	205±10*	201±9*	188±9*
■ 21	252±7	243±14*	242±14*	233±15*
	Maternal body weight gain (grams)			
Gestation days:				
■ 6-8	4±4	1±4*	-3±4*	-10±5*
■ 9-11	10±3	8±3	6±3*	2±3*
■ 12-15	13±6	12±1	11±3	10±3
■ 16-20	40±8	38±6	41±8	46±9*
■ 6-20 (Total)	66±8	59±10*	55±11*	48±11*
	Maternal liver weight, gestation day 21			
■ **Absolute (grams)**	9.84±0.41	9.29±0.71*	9.41±0.76*	9.05±0.85*
■ **Relative** [a]	3.91±0.14	3.83±0.24	3.89±0.31	3.88±0.29
	Maternal kidney weight, gestation day 21			
■ **Absolute (grams)**	1.40±0.12	1.38±0.14	1.32±0.11	1.38±0.14
■ **Relative** [a]	0.56±0.05	0.57±0.06	0.55±0.04	0.59±0.06*

* Dunnett's test, α=0.05.

[a] Grams organ weight / 100 grams body weight

Table III. 29 Food and water consumption in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (John *et al.*, 1983)

	Telone (ppm)			
	0	**20**	**60**	**120**
	Food consumption (grams/day)			
Gestation days:				
■ 6-8	13±1	12±1*	10±1*	7±2*
■ 9-11	14±1	13±1*	12±1*	9±2*
■ 12-14	15±1	14±1*	13±1*	11±1*
■ 15-17	17±1	14±1*	15±1*	15±2*
■ 18-20	17±1	15±1*	16±1*	17±1*
	Water consumption (grams/day)			
Gestation days:				
■ 6-8	20±3	20±4	31±3	16±3*
■ 9-11	23±2	22±2*	23±3	20±4*
■ 12-14	26±2	25±3	27±3	26±3
■ 15-17	31±2	27±4*	30±4	34±5*
■ 18-20	29±2	26±3*	28±3	29±3

* Dunnett's test, α=0.05.

Table III. 30 Embryo and fetal parameters in pregnant Fischer 344 rats exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (John *et al.*, 1983)

Parameter	Telone (ppm) 0	20	60	120
# Bred	30	30	30	30
# Maternal deaths	0	0	0	0
% Pregnant	93 (28/30)	83 (25/30)	70 (21/30)*	80 (24/30)
# Litters	27	25	20	24
Corpora lutea / dam	11±1	11±1	11±1	11±1
Implantation sites / dam	10±2	11±2	10±2	10±3
% Preimplantation loss	8±13	9±15	12±17	11±21
Fetuses / litter	10±2	10±2	9±2	9±3
Resorptions / litter	0.3±0.6	0.6±0.8	0.5±0.5	0.5±0.8
% Implantations resorbed	3 (9/273)	6 (15/262)	6 (11/197)	5 (11/234)
% Litters with resorptions	30 (8/27)	44 (11/25)	55 (11/20)	33 (8/24)
# Litters totally resorbed	0	0	0	0
Resorptions / litters with resorptions	1.1 (9/8)	1.4 (15/11)	1.0 (11/11)	1.4 (11/8)
Dead fetuses	0	0	0	0
% males / females	53/47	48/52	52/48	32/57
Fetal weight (g)	4.38±0.17	4.26±0.24*	4.36±0.17	4.36±0.27
Fetal crown-rump length (mm)	43.4±2.1	44.9±2.9	44.0±3.2	44.0±3.0
Delayed ossification of vertebral centrum				
■ # Fetuses	8/264	8/245	10/186	14/221
(% Fetuses)	(3)	(3)	(5)	(6)
■ % Litters	6/27	7/25	7/20	12/24
(% Litters)	(22)	(28)	(35)	(50)

*Different from control, α=0.05.

3. Rabbit, preliminary

Kloes (1983) conducted a preliminary whole-body inhalation study in pregnant Fischer 344 rats and New Zealand White rabbits with a purpose of determining the maximum tolerated dose of Telone II [9] for developmental toxicity in both species. This summary concerns only the rabbit phase of the study. Dose groups consisting 7 pregnant rabbits were exposed to this test article at target concentrations of 0, 50, 150 or 300 ppm for 6 hr/day on gestation days 6-18. These corresponded to analytically determined concentrations of 0, 50±1, 148±8 and 301±3 ppm. Telone atmospheres were created by metering known amounts of liquid Telone into glass tubes followed by removal and vaporization with compressed air heated to 100°C, then dilution and mixing before introduction into the exposure chambers. Observations for toxic signs and mortality were conducted daily. Body weights were determined on gestation days (gd) 6, 9, 12, 15 and 19. Food and water consumption were not monitored. Rabbits were sacrificed on gd 19. Gross pathologic changes were noted immediately upon sacrifice. Necropsies included determinations of liver and kidney weights, number of implantations and resorbed fetuses, and early resorptions. Histopathology was not conducted.

Maternal effects

Mortality and clinical signs. Six of seven 300-ppm does were either found dead or sacrificed moribund within 24 hr of showing rear limb ataxia, decreased or no righting reflex and flaccid hind limb muscles. These signs were evident by gestation days (gd) 14-19. The remaining 300-ppm animal was not pregnant, leaving no animals for analysis of pregnancy or developmental effects. Neither deaths nor overt clinical signs were noted at 50 or 150 ppm. One 150-ppm doe died of bacterial pneumonia that the investigators considered unrelated to treatment.

Maternal body weights. Statistically significant body weight decrements were noted at 150 ppm on gd 15 and 19 (Table III.31). No statistically significant decrements in weight gain were noted, though mean weight losses were consistently observed at 150 ppm. Rabbits do not tend to gain appreciable weight during pregnancy, so the lack of a statistical decrement at 150 ppm was not surprising.

Kidney and liver weights. No statistically significant effects were observed on mean kidney and liver weights through 150 ppm (no 300-ppm animals survived to analysis), though it is noted that liver weights were somewhat less than controls at 150 ppm (Table III.31). In view of the large standard deviations in these data, the toxicologic significance of this reduction was not clear.

Gross pathology. No gross pathologic lesions related to Telone exposure were noted at 50 or 150 ppm (Table III.32). Among the 300-ppm does that were found dead or sacrificed moribund, lung congestion and/or edema were noted in 2/6, cloudy or mucoid middle ear exudates in 4/6, and mucoid exudate of the external nares in 2/6. A test article relation was not clear for these findings.

[9] 47.7% cis and 42.2% trans 1,3-D with the following impurities: 1.8% epichlorohydrin, 0.2% 2,3-dichloropropene, 1.4% 1,2-dichloropropane, 1.5% 1,3-dichloropropane and 2.2% 1-chlorohexane.

Embryo and fetal effects

No differences in reproductive parameters were noted through 150 ppm (Table III.33). There were no data available for the 300-ppm dose group due to mortality.

A maternal NOEL was established at 50 ppm based on body weight decrements at 150 ppm. A developmental NOEL was greater than 150 ppm (due mainly to mortality, no pregnancies were analyzed at 300 ppm). These NOELs were considered to be preliminary, as the study was intended to establish a maximum tolerated dose. The maternal body weight decreases were noted after 3 days of exposure and thus may be pertinent to the evaluation of potential risks arising from acute or short term exposures. This study was deemed supplemental.

Table III. 31 Body weights and organ weights in pregnant New Zealand White rabbits exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

	Telone (ppm)			
	0	**50**	**150**	**300**
	Maternal body weight (grams)			
Gestation day:				
■ 6	4480±434	4217±276	4156±355	n/a [a]
■ 8	4460±410	4198±348	4102±301	n/a
■ 12	4411±367	4201±308	4059±290	n/a
■ 15	4470±333	4231±268	4021±294*	n/a
■ 19	4485±371	4234±296	3956±324*	n/a
	Maternal body weight gain (grams)			
Gestation days:				
■ 6-9	-20±106	-19±98	-54±138	n/a [a]
■ 9-12	-51±221	3±96	-43±72	n/a
■ 12-15	59±53	31±86	-38±65	n/a
■ 15-19	15±16	3±85	-67±71	n/a
■ 6-19 (Total)	18±123	17±56	-202±236	n/a
	Maternal liver weight, gestation day 19			
■ **Absolute (grams)**	115.26±26.96	109.37±11.88	93.36±17.84	n/a
■ **Relative** [b]	2.58±0.66	2.59±0.30	2.36±0.37	n/a
	Maternal kidney weight, gestation day 19			
■ **Absolute (grams)**	16.31±5.10	17.13±2.03	16.42±1.68	n/a
■ **Relative** [a]	0.37±0.12	0.41±0.06	0.42±0.04	n/a

* Dunnett's test, α=0.05

[a] Insufficient animals were available for analysis at this dose.

[b] Grams organ weight / 100 grams body weight

Table III. 32 Gross pathologic observations in pregnant New Zealand White rabbits exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

	Telone (ppm)			
	0	**50**	**150**	**300**
Surviving animals				
Lungs ■ Consolidation, left diaphragmatic lobe, focal	0/7	0/7	1/6	0/1
External & skin ■ Exudate, mucoid, external nares	0/7	0/7	0/6	1/1
Decedents				
Lungs				
■ Congestion	n/a	n/a	0/1	2/6
■ Edema	n/a	n/a	0/1	2/6
■ Consolidation	n/a	n/a	1/1	1/6
Tongue ■ Tumor-like moss or nodule	n/a	n/a	0/1	1/6
External & skin				
■ Alopecia, inguinal	n/a	n/a	0/1	1/6
■ Exudate, cloudy or mucoid, middle ear, bilateral	n/a	n/a	0/1	4/6
■ Exudate, mucoid, external nares	n/a	n/a	1/1	2/6
Thoracic cavity ■ Adhesions, pleural cavity	n/a	n/a	1/1	0/6
Uterus ■ Pregnant	n/a	n/a	0/1	5/6

Table III. 33 Embryo and fetal parameters in pregnant New Zealand White rabbits exposed to Telone II by inhalation, 6 hr/day, gestation days 6-15 (Kloes *et al.*, 1983)

Parameter	Telone (ppm) 0	50	150	300
# Bred	7	7	7	7
# Maternal deaths	0	0	1	6
% Pregnant	86 (6/7)	100 (7/7)	86 (6/7)	71 (5/7)
# Litters	6	7	6	0
Corpora lutea / dam	11±3	12±2	10±2	n/a [a]
Implantation sites / dam	8±4	10±2	8±2	n/a
% Preimplantation loss	31±28	12±16	19±15	n/a
Fetuses / litter	7±3	9±3	7±2	n/a
Resorptions / litter	0.8±1.2	1.1±1.9	0.8±1.0	n/a
% Implantations resorbed	11 (5/47)	11 (8/71)	11 (5/47)	n/a
% Litters with resorptions	50 (3/6)	43 (3/7)	50 (3/6)	n/a
# Litters totally resorbed	0	0	0	n/a
Resorptions / litters with resorptions	2.7 (8/3)	1.7 (5/3)	1.7 (5/3)	n/a

*Different from control, α=0.05

[a] Six of 7 animals died before the end of gestation. The remaining animal was not pregnant

4. Rabbit, complete

John (1983) exposed 25 - 31 inseminated female New Zealand White rabbits to Telone II (90.1% 1,3-D, consisting of 47.7% trans and 42.4% cis isomers) by the inhalation route for 6 hr/day on gestation days (gd) 6-18. The study was conducted to identify possible effects on developmental processes during the period of major organogenesis in the rabbit. The day of insemination was designated as gd 0. Inhalation exposures occurred in whole-body chambers at target concentrations of 0, 20, 60 or 120 ppm, which, like the parallel rat developmental study (John, 1983), were created by directing metered quantities of Telone into vaporization tubes, followed by removal of the tube contents into the chamber with a preheated compressed air stream. Mean analytical concentrations, determined once/hr by spectrophotometry, were essentially equivalent to the target concentrations. Standard observations in the does were made for mortality / moribundity, body weights (gestation days 6, 9, 12, 15, 19 and 29) and maternal liver and kidney weights (time of sacrifice). Food and water consumption was not monitored. Rabbits were euthanized on gd 29. Pregnancy observations included number and, where applicable, position of fetuses *in utero*, live and dead fetuses, implantation sites, resorption sites and corpora lutea. Fetuses were examined for gender, weight, crown-rump length, gross external pathology, soft tissue alterations and skeletal alterations.

Maternal effects

Mortality / moribundity. There were three maternal deaths during treatment, including two that succumbed to pneumonia (control and 60 ppm) and one for which the cause of death was not evident upon necropsy (120 ppm).

Body weight. There may have been an effect of Telone on body weight gain during the first 3 days of exposure at 60 and 120 ppm, though the effect was at best equivocal and was, at any rate, not maintained after this time (Table III.34).

Organ weights. No treatment effects were observed on liver or kidney weights (absolute and relative).

Toxicity to embryos or fetuses

No overt embryo or fetal toxicity was observed (Table III.35). However, a small number of fetuses (1 at 60 ppm and 2 at 120 ppm) displayed delayed ossification of metacarpals or phalanges. These particular fetuses were smaller than their litter mates (12.3-27.7 grams *vs.* a control mean of 36.1 grams) and displayed generally delayed skeletal development. It is plausible that the slightly delayed maternal growth at these doses was the ultimate cause, though these effects were not deemed sufficient to establish a developmental LOEL.

The NOEL for maternal effects was 20 ppm based on the possible dose-dependent body weight decrements noted at 60 and 120 ppm. It should be noted that body weight decreases were noted by gd 8, and thus may be pertinent to the evaluation of potential risks arising from acute or short

term exposures. A NOEL for developmental effects was >120 ppm, as no clear developmental effects were noted in the study.

This study was acceptable according to FIFRA guidelines.

Table III. 34 Body weights and organ weights in pregnant New Zealand White rabbits exposed to Telone II by inhalation, 6 hr/day, gestation days 6-18 (John *et al.*, 1983)

	Telone (ppm)			
	0	**20**	**60**	**120**
	Maternal body weight (grams)			
Gestation day:				
■ 6	3984±337	3769±229	4072±359	3919±337
■ 9	4020±339	3781±194	4052±351	3852±451
■ 12	4075±334	3816±213*	4096±349	3965±424
■ 15	4155±332	3923±286	4151±323	3950±374
■ 19	4186±341	3937±254	4154±344	3986±396
■ 29	4303±341	4047±328*	4291±345	4228±335
	Maternal body weight gain (grams)			
Gestation days:				
■ 6-8	36±102	12±91	-37±61*	-67±375
■ 9-11	54±81	35±56	61±55	113±287
■ 12-14	80±66	107±135	56±169	-16±213
■ 15-18	43±63	14±124	3±157	36±229
■ 19-28	103±195	110±128	137±109	242±256*
■ 6-29 (Total)	319±206	278±200	218±138	309±235
	Maternal liver weight, gestation day 29			
■ **Absolute (grams)**	119.94±12.85	100.59±14.40	100.77±17.33	111.38±17.72
■ **Relative** [a]	2.61±0.29	2.48±0.21	2.36±0.42*	2.63±0.34
	Maternal kidney weight, gestation day 29			
■ **Absolute (grams)**	17.17±1.38	15.88±1.40*	16.67±1.67	17.41±1.88
■ **Relative** [a]	0.40±0.05	0.39±0.03	0.39±0.04	0.41±0.03

* Dunnett's test, α=0.05.

[a] Grams organ weight / 100 grams body weight

Table III. 35 Embryo and fetal parameters in New Zealand White rabbits exposed to Telone II by inhalation, 6 hr/day, gestation days 6-18 (John *et al.*, 1983)

Parameter	Telone (ppm) 0	20	60	120
# Bred	29	25	31	25
# Maternal deaths	1	0	1	1
% Pregnant	86 (25/29)	72 (18/25)	68 (21/31)	88 (22/25)
# Litters	24	18	17	21
Corpora lutea / dam	10±2	10±2	10±2	10±2
Implantations sites / doe	9±3	8±3	8±3	8±3
% Preimplantation loss	10±18	16±20	17±18	18±24
Fetuses / litter	8±2	8±3	7±3	8±3
Resorptions / litter	0.8±0.8	0.6±0.9	0.8±1.1	0.7±1.3
% Implantations resorbed	8 (18/218)	7 (10/147)	10 (14/135)	9 (15/174)
% Litters with resorptions	50 (12/24)	39 (7/18)	53 (9/17)	43 (9/21)
# Litters totally resorbed	0	0	1	0
Resorptions / litters with resorptions	1.5 (18/12)	1.4 (10/7)	1.6 (14/9)	1.7 (15/9)
Dead fetuses	0	0	0	0
% males / females	54 / 46	56 /44	45 / 55	49 / 51
Fetal weight (g)	37.13±5.02	37.66±4.97	36.69±4.60	36.10±6.86
Fetal crown-rump length (mm)	96.88±5.04	95.65±5.62	96.22±6.34	94.95±6.72
Delayed ossification of metacarpals or phalanges				
■ % Fetuses	0	0	1 (1)	2 (3)
■ % Litters	0	0	7 (1)	15 (3)

*Different from control, α=0.05.

Table III. 36 Summary of NOEL and LOEL values from reproductive and developmental inhalation studies on 1,3-D

Study type	NOEL	LOEL	Determining sign(s)	*Acceptability* Reference
Reproductive toxicity				
■ Rat **■ 2 generations** **■ 0, 10, 30 & 90 ppm**	Maternal: 30 ppm Reproductive: >90 ppm	Maternal: 90 ppm Reproductive: >90 ppm	Maternal: ↓ BW, nasal histopathology Reproductive: n/a	*Acceptable* (Breslin *et al.*, 1987)
Developmental toxicity				
■ Rat preliminary **■ gd 6-15** **■ 0, 50, 150 & 300 ppm**	Maternal: <50 ppm Dvpmtl.: 150 ppm	Maternal: 50 ppm Dvpmtl.: 300 ppm	Maternal: BW decrements Dvpmtl.: Resorptions	*Supplemental* (Kloes *et al.*, 1983)
■ Rat complete **■ gd 6-15** **■ 0, 20, 60 & 120 ppm**	Maternal: <20 ppm Dvpmtl.: 60 ppm	Maternal: 20 ppm Dvpmtl.: 120 ppm	Maternal: BW & food consumption decrements Dvpmtl.: delayed ossification	*Acceptable* (John *et al.*, 1983)
■ Rabbit preliminary **■ gd 6-18** **■ 0, 50, 150 & 300 ppm**	Maternal: 50 ppm Dvpmtl.: 150 ppm	Maternal: 150 ppm Dvpmtl.: 300 ppm	Maternal: BW decrements Dvpmtl.: mortality	*Supplemental* (Kloes *et al.*, 1983)
■ Rabbit complete **■ gd 6-18** **■ 0, 20, 60 & 120 ppm**	Maternal: 20 ppm Dvpmtl.: >120 ppm	Maternal: 60 ppm Dvpmtl.: >120 ppm	Maternal: BW decrements Dvpmtl.: no effects	*Acceptable* (John *et al.*, 1983)

IV. RISK ASSESSMENT

A. Hazard identification

1. Acute / short-term toxicity

Acute or short term inhalation exposure to high concentrations of 1,3-D is known to result in upper respiratory symptoms in humans, including chest tightness, irritated and watery eyes, dizziness and runny nose. Laboratory rats exposed for 4 hours to 1,3-D vapors at doses of 300 ppm and higher exhibited labored, slow or exaggerated breathing; liver and lung congestion; lung edema; hydrothorax; corneal opacity; closed eyelids; visceral congestion; hunched posture; restlessness; pawing behavior; and body weight losses or body weight gain reductions (Cracknell *et al.*, 1987; Streeter *et al.*, 1987; Nitschke *et al.*, 1990a) Death was often observed above 600 ppm.

Data on clinical or pathologic signs were not adequate in strictly acute inhalation studies to set regulatory NOELs and LOELs, as the high dose ranges used in those studies were designed to determine LC_{50} values. Consequently, this information was garnered from body weight measurements taken in early time points (*i.e.*, up to 13 consecutive days, but usually one week or less) in ten inhalation subchronic, chronic and developmental toxicity studies.

The most common and sensitive effects occurring in these studies was a reduction in body weight and/or body weight gain, observed in rats, mice and rabbits. Virtually no other clinical signs or pathologies were noted at the dose ranges employed. The body weight effects, which were likely generalized expressions of animal stress, occurred early in the treatment period (*i.e.*, within 1-13 days) and continued after repeated exposures at the higher end of the concentration ranges employed. Similar concentrations of 1,3-D in different species elicited comparable decrements in body weight or weight gain.

a. Benchmark concentration modeling of the 1,3-D-induced body weight decrements

Benchmark concentration (BMC) modeling was used to determine the critical point of departure for the acute / short term body weight effect. BMC is a method by which a threshold---or benchmark concentration---is established for a toxicologic endpoint using mathematically fitted curves to model the data. The BMC approach uses most or all of the dose range and is applicable even in cases in which the relevant effect was observed at the lowest dose, *i.e.*, when a NOEL was not observed. This approach is widely considered to minimize uncertainties in toxicity threshold determinations. The operative value in BMC modeling is the BMCL, or the lower 95^{th} percentile confidence limit on the effective concentration (*i.e.*, the BMC) necessary to induce the response.

USEPA's Benchmark Dose Software version 2.6.0. (http://www.epa.gov/ncea/bmds/) was used to calculate BMCLs for 1,3-D (USEPA, 2012b). These values were derived from short term measurements of body weights in rats and mice as reported in the inhalation toxicity studies in

Table IV.1 below[10]. A benchmark response (BMR) of one standard deviation (SD, 1σ), was used to generate the $BMC_{1\sigma}$ as well as a $BMCL_{1\sigma}$.

Among the several available algorithms, the exponential, linear and polynomial algorithms provided the best observed curve fits. This was based on (a) statistical tests for goodness-of-fit, (b) lowest Akaike Information Criteria (AIC) score, (c) visual inspection of the curve and (d) the magnitude of the residuals (see Appendices I and II).

The estimated acute inhalation $BMCL_{1\sigma}$ values ranged between 6 and 66 ppm for the six rat and mouse studies with statistically analyzable data (Table IV.1), with the lowest value of 6 ppm resulting from analysis of body weight decrements in female rats occurring during the first week of a 90-day subchronic mouse study (Coate, 1979). However, the latter data were not used to characterize acute / short term risk, as they were considered too far outside the narrow range established in the other five studies. In addition, the purity of the test article used by Coate was not characterized, prohibiting calculation of an HEC. Instead, the 13-week rat study of Stott *et al*. (Stott *et al.*, 1984), which established a BMCL of **49 ppm** based on weight decrements in males at 3 days, was used for this purpose. This study was chosen because the 3-day time period more closely approximated an acute exposure regimen than the other studies.

[10] Note: body weight data from rabbit studies were also examined for this purpose. However, as noted in Table IV.1, those data could not be modeled by the available algorithms.

Table IV. 1 Benchmark concentration values based on body weight decreases after short term inhalation exposure of rats and rabbits to 1,3-D

Study	Air concentrations (ppm)	Exposure Duration (days)	$BMC_{1\sigma}$ (ppm)	$BMCL_{1\sigma}$ (ppm)
■ Rat ■ Dominant lethal ■ (Gollapudi *et al.*, 1997)	0, 10, 60, 150	7	82[b]	66[b]
■ Rat ■ 2-year chronic ■ (Lomax *et al.*, 1987)	0, 5, 20, 60	6	59[d]	53[d]
■ Rat ■ 13-week subchronic ■ (Stott *et al.*, 1984)	0, 10, 30, 90, 150	3	(♂/♀) 64/68[c]	(♂/♀) 49/51[c]
■ Rat ■ 13-week subchron.ic ■ (Coate, 1979)	0, 10, 30, 90	7	(♀) 19	(♀) 6
■ Rat ■ Developmental ■ (John *et al.*, 1983)	0, 20, 60, 120	4	79[c]	61[c]
■ Mouse ■ 2-year chronic ■ (Stott *et al.*, 1987)	0, 5, 20, 60	7	(♂/♀) 58 /51[b]	(♂/♀) 44/40[b]
■ Rabbit ■ Developmental ■ (John *et al.*, 1983)	0, 20, 60, 120	4	a	a
■ Rat ■ Developmental ■ (Kloes *et al.*, 1983)	0, 50, 150, 300	5	a	a
■ Rabbit ■ Developmental ■ (Kloes *et al.*, 1983)	0, 50, 150, 300	5	a	a

[a] Data could not be modeled by the available algorithms

[b] BMDS 2.6.0 Exponential mode

[c] BMDS 2.6.0 Linear model

[d] BMDS 2.6.0 Polynomial modelA 1 SD effect level, downward adverse direction, and constant variance were used to generate a family of continuous data models for each dataset (USEPA, 2012b).

See Section III (*Toxicology Profile*) for bodyweight data used to generate the above BMC(L) values.

b. Calculation of Human Equivalent Concentrations (HECs) using the Regional Gas Dose Ratio (RGDR) approach---short term exposure

Human equivalent concentrations (HECs) were estimated after calculating regional gas dose ratios (RGDRs). RGDRs are multipliers that are applied to the air concentrations used in the laboratory animal experiments in order to account for physiologic differences between animal and human respiration. The precise form of the RGDR equation depends on the likely route of toxicologic action, which is either systemic---in which case the relative distribution of the compound between vapor and blood is the essential determinant---or portal of entry, where the minute volume-to-surface area ratio in animals divided by that same ratio in humans is the determinant. The HEC calculations for 1,3-D assumed the former (*i.e.*, systemic) for the short term body weight effect and the latter (*i.e.*, portal of entry) for the subchronic and chronic nasal respiratory effect (see below). For oncogenesis, calculations were executed for both scenarios.

The equation for the HEC is as follows:

$$HEC = \text{exptl. concentration} \times (D_a / D_h) \times (W_a / W_h) \times RGDR$$

D_a: duration of animal exposure (hr/day)
D_h: duration of anticipated human exposure (hr/day)
W_a: duration of animal exposure (days/wk)
W_h: duration of anticipated human exposure (days/wk)

The RGDR methodology adopted in this assessment is described in detail in a review of inhalation reference concentration methodology by USEPA (1994) and in more abbreviated fashion in USEPA's 2007 risk assessment on 1,3-D (USEPA, 1994; USEPA, 2007). The relevant section of the latter document is quoted here for systemic effects (p. 88), with caveats noted in the Risk Appraisal section (section V.):

> In the case of systemic effects, the RGDR is defined as the ratio of the blood:gas partition coefficient of the chemical for the test species to humans ($H_{b/g\ animal}/H_{b/g\ human}$). When this ratio is unknown or when the $H_{b/g\ animal} > H_{b/g\ human}$ a default value of 1.0 is used as the RGDR. This default is based on the observation that for chemicals where partition coefficient data are available in both rats and humans the RGDR value has usually been comparable or slightly higher than 1. Thus, the use of an RGDR of 1 results in a protective calculation of the inhalation risk. Some of the key assumptions fundamental to the use of the RfC methodology to derive a HEC based on systemic effects include:
>
> 1) all the concentrations of inhaled gas within the animal's body are periodic with respect to time (i.e. periodic steady state - the concentration vs time profile is the same for every week). Periodicity must be attained for at least 90% of the exposure.
>
> 2) in the respiratory tract, the air, tissue, capillary blood concentration are in equilibrium with respect to each other.

3) systemically, the blood and tissue concentrations are in equilibrium with respect to each other.

In the case of 1,3-dichloropropene, the physicochemical properties and metabolism data for the compound indicate that these conditions (i.e. periodicity and equilibrium between different compartments) will be achieved in a very short period of time. Under these conditions, therefore, the use of the RfC methodology to estimate acute inhalation risk is appropriate.

In the absence of precise blood:gas ratio data on 1,3-D for humans or laboratory animals, the systemic RGDR defaulted to 1 on the assumption that the actual RGDR was greater than 1 (USEPA, 1994). Calculation of the HEC for the critical mouse study of Stott, 1984 thus reduced to an adjustment of the BMCL for exposure time, as noted below for both non-occupational and occupational exposure (Stott *et al.*, 1984):

Non-occupational

$HEC = POD \times (\text{formulation purity}) \times (D_a / D_h) \times (W_a / W_h) \times RGDR$

$HEC = 49 \text{ ppm} \times (0.91) \times (6 \text{ hr} / 24 \text{ hr}) \times (3 \text{ days} / 3 \text{ days}) \times 1 = 11.15 \approx$ **11 ppm**

Occupational

$HEC = POD \times (\text{formulation purity}) \times (D_a / D_h) \times (W_a / W_h) \times RGDR$

$HEC = 49 \text{ ppm} \times (0.91) \times (6 \text{ hr} / 8 \text{ hr}) \times (3 \text{ days} / 3 \text{ days}) \times 1 = 33.44 \approx$ **33 ppm**

These HECs are included in Tables IV.4 and IV.5 below, along with the HECs derived from the other studies for which weight decrement BMCLs were calculated. In the absence of age-specific acute body weight data, infant / child HECs were the same as for adults, though an additional database uncertainty factor of 3 was included. [11]

2. Subchronic / seasonal toxicity

The critical inhalation LOEL for the evaluation of seasonal exposure risks was 30 ppm (Table III.11). This was based on the appearance of of "very slight" hyperplasia of the nasal respiratory epithelium in the 13-wk rat study of Stott *et al*. (Stott *et al.*, 1984) in 2/10 males at 30 ppm. At the mid dose of 90 ppm, 10/10 males and 10/10 females exhibited hyperplasia of the respiratory epithelium, also graded as "very slight". By 150 ppm all animals displayed "slight" hyperplasia plus "slight" degeneration of the olfactory epithelium. The increased incidence, severity and number of respiratory system effects with increasing air concentration attested to their reliability as LOEL determinants. Bladder pathology was also observed in the mouse subchronic and chronic inhalation studies at similar air concentrations (Stott *et al.*, 1984; Stott *et al.*, 1987) though this was not used to characterize seasonal risk because combination of the BMCL10 of 16 ppm (see below) with the extrathoracic RGDR of 0.115 resulted in an HEC of 0.30 ppm,

[11] An uncertainty factor of 3 was adopted due to the relative mildness of the weight decrement in the vicinity of the BMCL concentrations.

much lower than what would result from use of a systemic endpoint (*eg*., bladder histopathology) with its RGDR of 1.

Coate made similar observations of nasal histopathology at 32 ppm in a prior rat subchronic inhalation study. Incidence of histopathologic signs in females was 0/10, 0/10, 9/10 and 10/10 at 0, 12, 32 and 93 ppm, respectively, for decreased nasal epithelial cytoplasm and 0/10, 0/10, 8/10 and 10/10 for disorganization of nuclei, resulting in a NOEL/LOEL of 12/32 ppm (Coate, 1979). The subchronic NOEL/LOEL of 30/90 ppm in mice---based on body weight decrements, organ weight changes, nasal histopathology and urinary bladder histopathology (Stott *et al.*, 1984)---supported the critical subchronic LOEL designation. [12]

a. Benchmark concentration modeling: subchronic end-points

As with the acute bodyweight decrements, BMC modeling was used to determine the critical point of departure for subchronic effects. Incidence data for nasal histopathology in rats from the two studies described in the previous paragraph was modeled using a benchmark response rate of 10% to generate the BMC_{10} and $BMCL_{10}$ values.

Among the several available algorithms for quantal data, the gamma and log-logistic algorithms provided the best observed curve fits. This was based on (a) statistical tests for goodness-of-fit, (b) lowest Akaike Information Criteria (AIC) score, (c) visual inspection of the curve and (d) the magnitude of the residuals (see Appendix III).

The estimated subchronic inhalation $BMCL_{10}$ values ranged from 9 to 16 ppm for the two rat studies with statistically analyzable data (Table IV.2), with the two lowest values of 9 ppm resulting from analysis of incidences of decreased cytoplasm and disorganization of nuclei in nasal epithelial cells occurring during a 90 day subchronic mouse study (Coate, 1979). However, the 16 ppm $BMCL_{10}$ value from the Stott study was used to calculate the critical subchronic HEC because the purity of the test article used in the Coate study was not provided.

[12] It is also noted that a NOEL/LOEL of 30/90 ppm was established for body weight decrements in the rat study, an effect which was noted early (short term $BMCL_{1SD}$, ♂/♀, = 49/51 ppm) and maintained throughout the study.

Table IV. 2 Benchmark concentration values from subchronic inhalation toxicity studies with 1,3-D

Study	Air concentrations (ppm)	Toxicity end point	BMC_{10} (ppm)	$BMCL_{10}$ (ppm)
■ **Rat** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)**	0, 10, 30, 90, 150	Hyperplasia of nasal respiratory epithelium	(♂) 27	(♂) 16 [a]
■ **Rat** ■ **13-wk subchronic** ■ **(Coate, 1979)**	0, 10, 30, 90	Decreased cytoplasm in nasal epithelim	(♀) 24	(♀) 9 [b]
■ **Rat** ■ **13-wk subchronic** ■ **(Coate, 1979)**	0, 10, 30, 90	Disorganization of nuclei in nasal epithelium	(♀) 25	(♀) 9 [b]

[a] BMDS 2.6.0 Gamma and log-logistic model
[b] BMDS 2.6.0 Log-logistic model

b. Calculation of HEC using the RGDR approach---subchronic (seasonal) exposure

Histopathology of the nasal epithelium was, by definition, an extrathoracic, portal of entry-driven event. RGDR determinations for portal-of-entry effects were described in USEPA's 2008 updated risk assessment on 1,3-D (USEPA, 2008a). The relevant section---pp. 65-66---is quoted here:

> When the critical toxic effect in a study occurs in the respiratory tract (i.e port of entry effects), the RGDR is not related to the blood:gas partition coefficient of the compound but rather the ratio of the minute volume (MV) to the surface area (SA) of the affected region. In these instances, attaining periodicity or equilibrium between the compartments is not critical (since the effect is a function of the direct interaction between the inhaled compound and the affected region in the respiratory tract) and the RGDR may be calculated using the following equation:
>
> $$RGDR = (MV_{animal} / SA_{animal}) \div (MV_{human} / SA_{human})$$
>
> Where:
> MV_{animal}: Minute volume for the test species (varies depending on body weight)
> SA_{animal}: Surface area of the affected region in animals
> MV_{human}: Minute volume for humans (default value is 13.8 l/min)
> SA_{human}: Surface area of the affected region in humans
>
> The MV_{animal} is calculated using the allometric scaling provided in (USEPA, 1994).The equation for calculation of the MV_{animal} is:
>
> $$\ln MV_{animal} = b_0 + b_1 \ln(BW)$$

Where:
ln MV_{animal}: natural logarithm of the minute volume
b_0 : species specific intercept used in the algorithm to calculate minute volumes based on body weight
b_1: species specific coefficient used in the algorithm to calculate minute volumes based on body weight
ln BW: natural logarithm of the body weight (expressed in kg)

The values for the species-specific parameters used to calculate the MV_{animal} based on body weight and the values for the surface areas of various regions of the respiratory tract (extrathoracic, thoracic, and pulmonary) are provided in the EPA document "Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry" (USEPA, 2008a).

USEPA (1994) recommends default extrathoracic surface areas of 15 cm^2 (rat) and 200 cm^2 (human), with the rat minute volume calculated using the allometric scaling equation noted above. As extrathoracic surface areas for infants and children were not available, those RGDRs and HECs were not calculated.[13]

$RGDR = (MV_{rat} / SA_{rat}) \div (MV_{human} / SA_{human})$
MV_{rat}: $lnMV_{rat} = -0.578 + 0.821(lnBW)$
BW for male Fischer 344 rat (subchronic): 0.180 kg
BW for female Fischer 344 rat (subchronic): 0.124 kg
$lnMV_{rat\text{-}male} = -1.99$
$MV_{rat\text{-}male} = 0.137$ L/min
$lnMV_{rat\text{-}female} = -2.29$
$MV_{rat\text{-}female} = 0.101$ L/min
$SA_{et\text{-}rat}$: 15 cm^2
$SA_{et\text{-}human}$: 200 cm^2
$RGDR_{rat\text{-}male} = (0.137 \text{ L/min} / 15 \text{ cm}^2) \div (13.8 \text{ L/min} / 200 \text{ cm}^2) = 0.133$
$RGDR_{rat\text{-}female} = (0.101 \text{ L/min} / 15 \text{ cm}^2) \div (13.8 \text{ L/min} / 200 \text{ cm}^2) = 0.098$
$RGDR_{rat\text{-}mean} = (0.133 + 0.098) \div 2 = 0.115$

Non-occupational
HEC = (16 ppm) x (0.91) x (6 hr / 24 hr) x (5 days / 7 days) x (0.115) = **0.30 ppm**

Occupational
HEC = (16 ppm) x (0.91) x (6 hr / 8 hr) x (5 days / 7 days) x (0.115) = **0.90 ppm**

These HECs are included in Tables IV.4 and IV.5 below, along with the HEC derived from the subchronic mouse study (Stott *et al.*, 1984).

[13] Risk for these demographics was assessed by applying an additional uncertainty factor of 3 to the adult target MOE, as was done with short-term toxicity above.

3. Chronic / annual toxicity and oncogenicity

a. Chronic (non-oncogenic) toxicity

The critical inhalation LOEL for the evaluation of chronic exposure scenarios was **20 ppm** (Table III.18). This was established in the mouse 2-year inhalation study (Stott *et al.*, 1987), which showed hyperplasia of the nasal respiratory epithelium in females, hyperplasia and hypertrophy of the urinary bladder transitional epithelium in males and females, and roughened, irregular opaque urinary bladder surface in females at that dose. These signs increased in incidence and severity at the high dose of 60 ppm.

Support for the critical LOEL came from the rat chronic inhalation study of (Lomax *et al.*, 1987), though the rat value was based on decreased thickness and erosion of the olfactory epithelium in only 1/50 animals at 20 ppm. These signs increased in incidence at 60 ppm to 20/50 and 15/50, respectively, and included an additional sign: fibrosis of the olfactory submucosa in 6/50 animals. Body weight was also impacted at 20 ppm in males, though the reduction was not nearly so consistently observed as at the high dose.

b. Benchmark concentration modeling: chronic endpoints

As with the acute and subchronic end-points, BMC modeling was used to determine the critical point of departure for chronic effects. BMC values were derived from incidences of nasal histopathology in rats and mice. Table IV.3. provides the results in both species for the most sensitive (*i.e.*, lowest BMC and BMCL values) endpoints. A benchmark response of 10% was used to generate the BMC_{10} as well as the $BMCL_{10}$.

Among the several available algorithms for quantal data, the logistic, multistage, quantal-linear, and log-probit algorithms provided the best observed curve fits. As with the acute and subchronic endpoints, this was based on (a) statistical tests for goodness-of-fit, (b) lowest Akaike Information Criteria (AIC) score, (c) visual inspection of the curve and (d) the magnitude of the residuals (see Appendix IV).

$BMCL_{10}$ values ranged between 6 and 32 ppm (the later value came from modeling decreased thickness in the olfactory eptithelium in the Lomax rat study – it does not appear in Table IV.3.), with the lowest value of 6 ppm resulting from analysis of the incidence of bilateral hypertrophy and hyperplasia in the nasal respiratory mucosa of female mice occurring during the 2-year chronic toxicity mouse study (Stott *et al.*, 1987)

Table IV. 3 Benchmark concentration values from chronic inhalation toxicity studies with 1,3-D

Study	Air concentrations (ppm)	Toxicity end point	BMC_{10} (ppm)	$BMCL_{10}$ (ppm)
■ **Rat** ■ **2-yr chronic** ■ **(Lomax *et al.*, 1987)**	0, 5, 20, 60	Erosion of the olfactory epithelium	(♀) 34 [a]	(♀) 27 [a]
■ **Mouse** ■ **2-yr chronic** ■ **(Stott *et al.*, 1987)**	0, 5, 20, 60	Bilateral hypertrophy & hyperplasia of the nasal respiratory mucosa	(♀) 10 [b]	(♀) 6 [b]

[a] BMDS 2.6.0 multistage model
[b] BMDS 2.6.0 Log probit model

c. Calculation of HECs using the RGDR approach---chronic (annual) exposure

Both nasal respiratory and bladder lesions were LOEL determinants for chronic toxicity, suggesting both portal-of-entry and systemic routes of toxicity. However, HECs generated by a systemic approach, which by USEPA's convention assumes an RGDR of 1, are greater---thus less health-protective---than those generated by a portal-of-entry approach, with RGDRs much less than 1 (see footnote 15 and the following paragraphs). Consequently, the critical chronic HEC was calculated assuming portal-of-entry toxicology. [14]

The portal-of-entry effect was bilateral hypertrophy and hyperplasia of the nasal respiratory mucosa, an extrathoracic parameter requiring use of extrathoracic surface areas to calculate RGDRs. USEPA (1994) recommends a default extrathoracic surface area of 3 cm^2 for the mouse. The human extrathoracic surface area of 200 cm^2 noted above for subchronic toxicity also applied in the chronic case. Minute volume for the female B6C3F1 mouse under chronic exposure conditions was calculated using the allometric scaling equation designated in USEPA (1994) (USEPA, 1994).

$RGDR = (MV_{mouse} / SA_{mouse}) \div (MV_{human} / SA_{human})$

MV_{mouse}: $lnMV_{mouse} = 0..326 + 1.050(lnBW)$

BW for female B6C3F1 mouse (chronic): 0.0353 kg

$lnMV_{female} = -3.185$

$MV_{female} = 0.041$ L/min

$SA_{et\text{-}mouse}$: 3 cm^2

$SA_{et\text{-}human}$: 200 cm^2

$RGDR_{mouse\text{-}female} = (0.041 \text{ L/min} / 3 \text{ cm}^2) \div (13.8 \text{ L/min} / 200 \text{ cm}^2) = 0.198$

Non-occupational

HEC = (6 ppm) x (0.92) x (6 hr / 24 hr) x (5 days / 7 days) x (0.198) = **0.20 ppm**

Occupational

HEC = (6 ppm) x (0.92) x (6 hr / 8 hr) x (5 days / 7 days) x (0.198) = **0.59 ppm**

These HECs are included in Tables IV.4 and IV.5 below, along with the HEC derived from the chronic rat study.

[14] Non-occupational chronic HEC using a systemic RGDR of 1:
(6 ppm) x (6 hr / 24 hr) x (5 days / 7 days) x (1) x (0.92) = 0.99 ppm
^^^^^^^^^^^^^^^^^^^^

Occupational chronic HEC using a systemic RGDR of 1:
(6 ppm) x (6 hr / 8 hr) x (5 days / 7 days) x (1) x (0.92) = 2.96 ppm

Table IV. 4 HEC array for non-occupational scenarios

Study	LOEL NOEL (ppm)	BMCL (ppm)	$\frac{D_a}{D_h}$	$\frac{W_a}{W_h}$	RGDR	HEC (ppm)	Uncertainty factors Inter	 Intra	 Database
Short term – (RGDR calculation: systemic [a])									
■ **Rat** ■ **Dominant lethal** ■ **(Gollapudi *et al.*, 1998)** ■ **96% 1,3-D**	n/a	60	6 24	7 7	1	14	3	10	1-adult 3-child
■ **Rat** ■ **2-yr chronic** ■ **(Lomax *et al.*, 1987)** ■ **92% 1,3-D**	n/a	54	6 24	6 6	1	12	3	10	1-adult 3-child
■ **Rat** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **91% 1,3-D**	n/a	49	6 24	3 3	1	**11**	3	10	1-adult 3-child
■ **Rat** ■ **Developmental** ■ **(John *et al.*, 1983)** ■ **90% 1,3-D**	n/a	61	6 24	9 9	1	14	3	10	1-adult 3-child
■ **Mouse** ■ **2-yr chronic** ■ **(Stott *et al.*, 1987)** ■ **92% 1,3-D**	n/a	40	6 24	5 7	1	7	3	10	1-adult 3-child
Seasonal – (RGDR calculation: extrathoracic [b])									
■ **Rat** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **90.9% 1,3-D**	30 10	16	6 24	5 7	0.115 [c]	**0.30**	3	10	1-adult 3-child
■ **Rat** ■ **13-wk subchronic** ■ ■ **purity unknown**	32 12	9	6 24	5 7	0.115 [c]	NC [d]			
■ **Mouse** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **90.9% 1,3-D**	90 30	Not modeled [d]							
Annual – (RGDR calculation: extrathoracic [b])									
■ **Rat** ■ **2-yr chronic** ■ **(Lomax *et al.*, 1987)** ■ **92.1% 1,3-D**	20 5	34	6 24	5 7	0.245 [e]	1.37	3	10	1-adult 3-child
■ **Mouse** ■ **2-yr chronic** ■ **(Stott *et al.*, 1987)** ■ **92.1% 1,3-D**	20 5	6	6 24	5 7	0.198 [f]	**0.20**	3	10	1-adult 3-child

[a] Body weight effects were considered to occur through systemic toxicity. In the absence of chemical specific blood-:vapor partition data, the RGDR was set to a default of 1.

[b] Seasonal and annual toxicity were considered to result from portal-of-entry impacts on the extrathoracic sector of the respiratory tree. Inputs for minute volume and surface area used to calculate RGDRs for rats and mice appear in the following footnotes.

[c] RGDRs for male and female Fischer 344 rats, subchronic conditions, were calculated as shown in the text for this section. Rat minute volumes were 0.137 and 0.101 L/min for males and females, respectively, under subchronic exposure conditions. Rat extrathoracic surface area was 15 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

[d] HEC was not calculated for this study because the purity of the test article was not reported in the study.

[e] The mouse 13-wk subchronic study was not modeled because mice were notably less sensitive than rats with regard to extrathoracic toxicity as evidenced by their considerably higher NOELs and LOELs than rats.

[f] RGDR for male Fischer 344 rats, chronic conditions, were calculated as shown in the text for this section. Male rat minute volume was 0.254 L/min under chronic exposure conditions. Rat extrathoracic surface area was 15 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

[g] RGDR for female B6C3F1 mice, chronic conditions, were calculated as shown in the text for this section. Female mouse minute volume was 0.041 L/min under chronic exposure conditions. Mouse extrathoracic surface area was 3 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

Table IV. 5 HEC array for occupational scenarios

Study	LOEL NOEL (ppm)	BMCL (ppm)	$\frac{D_a}{D_h}$	$\frac{W_a}{W_h}$	RGDR	HEC (ppm)	Uncertainty factors Inter	 Intra	 Database
Short term – (RGDR calculation: systemic [a])									
■ **Rat** ■ **Dominant lethal** ■ **(Gollapudi *et al.*, 1998)** ■ **96% 1,3-D**	n/a	60	6 8	7 7	1	43	3	10	1-adult
■ **Rat** ■ **2-yr chronic** ■ **(Lomax *et al.*, 1987)** ■ **92% 1,3-D**	n/a	54	6 8	6 6	1	37	3	10	1-adult
■ **Rat** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **91% 1,3-D**	n/a	49	6 8	3 3	1	**33**	3	10	1-adult
■ **Rat** ■ **Developmental** ■ **(John *et al.*, 1983)** ■ **90% 1,3-D**	n/a	61	6 8	9 9	1	41	3	10	1-adult
■ **Mouse** ■ **2-yr chronic** ■ **(Stott *et al.*, 1987)** ■ **92% 1,3-D**	n/a	40	6 8	5 7	1	20	3	10	1-adult
Seasonal – (RGDR calculation: extrathoracic [b])									
■ **Rat** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **90.9% 1,3-D**	30 10	16	6 8	5 7	0.115 [c]	**0.90**	3	10	1-adult
■ **Rat** ■ **13-wk subchronic** ■ **(Coate, 1979)** ■ **purity unknown**	32 12	9	6 8	5 7	0.115 [c]	NC [d]			
■ **Mouse** ■ **13-wk subchronic** ■ **(Stott *et al.*, 1984)** ■ **90.9% 1,3-D**	90 30	Not modeled [e]							
Annual – (RGDR calculation: extrathoracic [b])									
■ **Rat** ■ **2-yr chronic** ■ **(Lomax *et al.*, 1987)** ■ **92.1% 1,3-D**	20 5	34	6 8	5 7	0.245 [f]	4.11	3	10	1-adult
■ **Mouse** ■ **2-yr chronic** ■ **(Stott *et al.*, 1987)** ■ **92.1% 1,3-D**	20 5	6	6 8	5 7	0.198 [g]	**0.59**	3	10	1-adult

[a] Body weight effects were considered to occur through systemic toxicity. In the absence of chemical specific blood-:vapor partition data, the RGDR was set to a default of 1.

[b] Seasonal and annual toxicity were considered to result from portal-of-entry impacts on the extrathoracic sector of the respiratory tree. Inputs for minute volume and surface area used to calculate RGDRs for rats and mice appear in the following footnotes.

[c] RGDRs for male and female Fischer 344 rats, subchronic conditions, were calculated as shown in the text for this section. Rat minute volumes were 0.137 and 0.101 L/min for males and females, respectively, under subchronic exposure conditions. Rat extrathoracic surface area was 15 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

[d] HEC was not calculated for this study because the purity of the test article was not reported in the study.

[e] The mouse 13-wk subchronic study was not modeled because mice were notably less sensitive than rats with regard to extrathoracic toxicity as evidenced by their considerably higher NOELs and LOELs than rats.

[f] RGDR for male Fischer 344 rats, chronic conditions, were calculated as shown in the text for this section. Male rat minute volume was 0.254 L/min under chronic exposure conditions. Rat extrathoracic surface area was 15 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

[g] RGDR for female B6C3F1 mice, chronic conditions, were calculated as shown in the text for this section. Female mouse minute volume was 0.041 L/min under chronic exposure conditions. Mouse extrathoracic surface area was 3 cm^2. Human minute volume and extrathoracic surface area were 13.8 L/min and 200 cm^2, respectively.

c. Oncogenicity

Male mice exposed to 1,3-D by the inhalation route for 2 years exhibited a statistically elevated incidence of bronchioloalveolar adenomas at a nominal air concentration of 60 ppm (22/50: 44%; Table III.16) (Stott *et al.*, 1987). The incidence rate at 20 ppm (13/49: 27%) was also higher than concurrent controls (9/49: 18%) and low dose animals,(6/50: 12%) though a treatment relation was not certain, as 7 previous chronic studies showed a historical control incidence range between 7 and 32%. However, in view of the apparent dose dependence and the evidence for genotoxicity , the linearized multistage cancer model (BMCS version 2.6) was used to characterize the dose response. The multistage model is considered standard for cancer bioassay modeling when there is no evidence for a more biologically based model and when the data are amenable to modeling, as is the case here (USEPA, 2012b). It is also used when there is evidence for genotoxicity, as is the case for 1,3-D. A benchmark response (BMR) of 10% "extra risk" was chosen to determine the slope potency. The resultant curve along with the BMR output appears below in Appendix V.

For multistage dose modeling, the air concentrations used in the mouse study were converted to human equivalent concentrations (HECs) assuming two different mechanistic scenarios: (1) adenomas arose following direct interaction of inspired 1,3-D with the tracheobronchial and pulmonary epithelial surfaces of the lung. This portal-of-entry scenario would be similar to the subchronic and chronic induction of nasal epithelial hyperplasia, but requiring a much higher RGDR to compute an HEC because the ratio of minute volume to involved respiratory system surface area was much less for humans than for mice; and (2) adenomas arose following absorption and circulatory redistribution to the lung of 1,3-D or its metabolites. As the second scenario invokes systemic exposure, dose scaling from mouse to human utilized a default RGDR of 1, similar to our treatment of acute toxicity. We chose to characterize lung tumorigenesis in both ways because the data did not point overwhelmingly to one or the other scenario, though we felt ultimately that the evidence tilted to the portal of entry scenario. The following observations were marshalled in support of portal of entry: (a) upper respiratory irritation occurred after acute, subchronic and chronic exposure in rodents (Cracknell *et al.*, 1987; Nitschke *et al.*, 1990b) and after acute exposure in humans (section III.B.1. above); in addition, rats decreased their breathing rate at 90 ppm (Stott and Kastl, 1986), which was interpreted as evidence for sensory irritation in the upper respiratory tract; (b) pharmacokinetic studies in rats showed definitively that inspired 1,3-D reaches the lower respiratory system (Stott and Kastl, 1986); (c) 1,3-D causes tumors on contact in other mouse tissues, including forestomach upon gavage exposure and skin (papillomas) upon dermal exposure (NTP, 1985); (d) skin sensitization resulted after dermal exposure in guinea pigs (Jeffrey, 1987); (e) oral, but not inhalation, exposure in rats caused liver adenomas, suggesting that local mechanisms were operative for liver tumors (Stott *et al.*, 1995). Supporting a systemic scenario is the following evidence: (a) 1,3-D is readily absorbed by the inhalation route in both rats (Stott and Kastl, 1986) and humans (Waechter *et al.*, 1992); (b) inhalation exposure leads to epithelial hyperplasia in the mouse bladder (Stott *et al.*,

1987) and, at higher concentrations, histopathologic changes in the kidneys, stomach and liver; (c) oral exposure in mice caused bronchioloalveolar tumors similar to those developing from inhalation exposure, suggesting that even by the inhalation route, absorption might be required for tumor development (NTP, 1985), though it is also possible that oral dosing led to inhalation exposure through reflux of volatilized or non-volatilized 1,3-D (Sells *et al.*, 2007; Damsch *et al.*, 2011a; Damsch *et al.*, 2011b).

HEC calculation, portal of entry scenario.

$RGDR = (MV_{mouse} / SA_{mouse}) \div (MV_{human} / SA_{human})$
MV_{mouse}: $\ln MV_{mouse} = 0..326 + 1.050(\ln BW)$
BW for male B6C3F1 mouse (chronic): 0.0373 kg
$\ln MV_{male} = -3.127$
$MV_{male} = 0.044$ L/min
SA_{mouse}: $SA_{tb\text{-}mouse} + SA_{pu\text{-}mouse} = 3.5 + 500 = 503.5$ cm^2
SA_{human}: $SA_{tb\text{-}human} + SA_{pu\text{-}human} = 3200 + 540{,}000 = 543{,}200$ cm^2
$RGDR_{mouse\text{-}female} = (0.044$ L/min / 503.5 cm$^2) \div (13.8$ L/min / 543,200 cm$^2) = 3.44$

HECs for each dose in the mouse study were calculated using the formulas indicated below. Separate HECs for resident / bystander / ambient scenarios and for occupational scenarios were developed due to the anticipated daily exposure times for these categories:

Resident / bystander / ambient:
HEC = (nominal dose) x (92% purity) x (6 hr / 24 hr) x (5 days / 7 days) x 3.44

Occupational:
HEC = (nominal dose) x (92% purity) x (6 hr / 8 hr) x (5 days / 7 days) x 3.44

HEC calculation, systemic scenario.

Resident / bystander / ambient:
HEC = (nominal dose) x (92% purity) x (6 hr / 24 hr) x (5 days / 7 days) x 1

Occupational:
HEC = (nominal dose) x (92% purity) x (6 hr / 8 hr) x (5 days / 7 days) x 1

The HECs and upper confidence limit air unit risk values appear below in Table IV.5. As noted above, the dose response curves and multistage model outputs for the portal of entry outputs

appear in Appendix V (systemic outputs are exactly the same, though multiplied by the 3.44x factor).

Table IV. 6 Human equivalent doses and incidence rates used to model the dose responsiveness of 1,3-D-induced bronchioloalveolar adenomas in male mice (Stott *et al.*, 1987)

Portal of entry scenario				
Nominal dose	**RGDR**	**HEC dose (resident-bystander-ambient)**	**HEC dose (occupational)**	**Incidence rate**
0 ppm	3.44	0 ppm	0 ppm	9/49 (18%)
5	3.44	2.83	8.48	6/50 (12%)
20	3.44	11.30	33.91	13/49 (27%)
60	3.44	33.91	101.73	22/50 (44%)
Air unit risk – upper confidence limit (ppm^{-1})		0.018	0.0059	n/a
Systemic scenario				
0 ppm	1	0 ppm	0 ppm	9/49 (18%)
5	1	0.82	2.46	6/50 (12%)
20	1	3.29	9.86	13/49 (27%)
60	1	9.86	29.57	22/50 (44%)
Air unit risk – upper confidence limit (ppm^{-1})		0.062	0.020	n/a

4. Genotoxicity

Based on the results of a series of *in vitro* and *in vivo* genotoxicity studies tests for this document, 1,3-D is considered to be genotoxic. In addition, several metabolites and degradates of 1,3-D exhibit genotoxic properties.

5. Reproductive toxicity

Reproductive parameters were not affected in the single inhalation reproductive toxicity study on 1,3-D available for review (Breslin *et al.*, 1987). For the purposes of this assessment, 1,3-D is not considered to be a reproductive toxicant by the inhalation route.

6. Developmental toxicity

There was no instance of fetal or developmental effects occurring at air concentrations lower than those that induced maternal toxicity in rats or rabbits (Kloes *et al.*, 1983); (John *et al.*, 1983). Consequently, 1,3-D is not considered to be a developmental toxicant by the inhalation route.

B. Exposure assessment

Exposure estimates are provided for scenarios representing handlers, reentry workers, and occupational and residential bystanders. The handlers consist of the fumigant applicator, the fumigant loader, and the tarp remover. 1,3-D is applied via shank, drip, and injection auger methods. During fumigation, the treated ground can be left open or covered in a tarp to trap the fumigant. Following the fumigation period, a worker enters the field and removes, cuts, or punches holes in the tarp to aerate the field. The bystander scenarios considered include the occupational bystander which could be adjacent to a field undergoing fumigation via shallow or deep shank without the use of a tarp, or via drip irrigation with the use of a tarp. In addition, 1,3-D breathing-zone air concentrations for the residential bystander located 100 feet from the edge of the treated field were estimated. A 100-foot buffer zone is mandated by CA permit conditions for any occupied structures (e.g., residences, schools), and, as a result, was applied when estimating exposure for the residential bystander. However, an occupational bystander could potentially be a field worker adjacent to the field being treated. There's no language in the CA permit conditions and certain product labels currently on DPR's active product list (i.e., Telone II, Telone EC, and Tri-Cal Trilone II), addressing this scenario. Hence, for the occupational bystander, the buffer zone was not incorporated into the exposure assessment. The estimates for the residential bystander were classified according to the potential source of 1,3-D (i.e., from the nearby treated field or from ambient air).

For each exposure scenario, the short-term and long-term breathing-zone air concentrations were estimated. The short-term air concentration (STAC) is defined as the daily 1,3-D breathing-zone air concentration of the worker (8-hr time-weighted-average or TWA) for up to one week, or the residential bystander (24-hr TWA) for up to one week. The long-term air concentration estimates consist of the seasonal, annual, and lifetime air concentrations or SAC, AAC, and LAC, respectively. The SAC is the daily (8-hr TWA for the worker, and 24-hr TWA for the residential bystander) 1,3-D breathing-zone air concentration anticipated for the use season in the highest use county. The AAC is the estimated 1,3-D air concentration to which a worker or bystander is exposed to throughout the year. For the occupational exposure scenarios, the LAC is the AAC multiplied by the assumed total number of years worked (40 years) divided by the assumed worker's lifetime of 75 years. For residential bystander exposures due to ambient 1,3-D air concentrations, multiple LAC estimates were generated for residents within a high-use area (Merced County).

The short- and long-term breathing-zone air concentration occupational exposure estimates were generated using four different sources of data. These sources consist of the 1,3-D breathing-zone air concentration data generated by the registrant (Houtman, 1993), surrogate chloropicrin air monitoring data (Beauvais, 2010), simulated air concentrations (Johnson, 2009a), and a 14-month 1,3-D ambient air monitoring study conducted by the registrant (Rotondaro and van Wesenbeeck, 2012b). In the cases of the handler scenarios, 1,3-D breathing-zone air concentrations were generated using registrant and surrogate data. The applicator (shallow shank

w/o tarp) scenario air concentrations were calculated using the registrant study. The shallow shank w/tarp, deep shank w/ and w/o tarp, drip, and injection auger applicator scenario air concentrations were generated using surrogate data. The loader scenario exposure estimates were generated using registrant data while the tarp remover exposure estimates were made using surrogate data. The reentry worker exposure estimates were generated using registrant data while the occupational bystander exposure estimates were calculated using computer simulated air concentrations and data from the ambient air monitoring study.

Residential bystander exposure estimates were generated using simulated air concentrations and air monitoring data. Simulated air concentrations were used to estimate short-term and seasonal breathing-zone air concentrations for residential bystanders at the edge of the buffer-zone (Powell, 2000; Johnson, 2009a; Johnson, 2009b). Air monitoring data were used to estimate STAC and.SAC values for residential exposure to ambient 1,3-D levels while simulated air concentrations were utilized to generate the AAC and LAC values for residential exposure to ambient 1,3-D levels (Table IV.7) (Rotondaro and Van Wesenbeeck, 2012a).

Table IV. 7 Data sources for exposure scenarios

Exposure Scenario	Worker Exposure Data	Simulated Data	Surrogate Data	Air Monitoring Data
applicator (shallow shank, w/o tarp) [a]	X			
applicator (shallow shank, w/ tarp) [b]			X	
applicator (deep shank w/ and w/o tarp) [b]			X	
applicator (drip w/ and w/o tarp) [b]			X	
applicator (injection auger) [b]			X	
loader [a]	X			
tarp remover [b]			X	
reentry worker [a]	X			
occupational bystander (shallow shank w/o tarp) [c]		X		X
occupational bystander (deep shank w/o tarp) [c]		X		X
occupational bystander (drip w/ tarp) [c]		X		X
residential bystander (exposure to 1,3-D at 100 ft from edge of treated field) [d]		X		
residential bystander (exposure to ambient levels of 1,3-D) [e]		X		X

[a] The applicator (shank, w/o tarp), loader, and reentry worker exposure estimates were derived from a registrant study investigating 1,3-D breathing-zone air concentrations for handlers loading and applying 1,3-D via the shank injection method, and for reentry workers (Houtman, 1993).

[b] No 1,3-D air concentration data were available for these exposure scenarios. A method utilizing surrogate chloropicrin air monitoring data for the tarp remover and applicator (shank w/tarp, deep shank w/ and w/o tarp, drip w/ and w/o tarp, and injection auger), and 1,3-D air monitoring data for the applicator (shank w/o tarp) was used to derive the exposure estimates for these scenarios.

[c] The occupational bystander (shallow shank, deep shank, and drip application method) short-term air concentrations, were generated via ISCST3 air concentration simulations (Johnson, 2009a). The worker is assumed to be located at the edge of the treated field during application for 8 hours. The air concentration utilized to generate this estimate was simulated at 3.04 meters from the edge of the field, the closest to the edge of the field in the simulation. The long-term air concentration estimates (seasonal, annual, and lifetime air concentrations, or SAC, AAC, and LAC values) were calculated using air monitoring data generated from 14 months of sampling in a 9-township area within Merced County (Rotondaro and van Wesenbeeck, 2012b).

[d] This residential exposure scenario represents exposure which occurs at the edge of the 100-foot buffer-zone of a field treated once annually. The short-term air concentration (STAC) and SAC were calculated using air concentrations simulated for 24-hr or 2-week flux generated for different application methods (shallow shank, deep shank, drip) (Johnson, 2009b; Johnson, 2009a), and for tree and vine crops (Powell, 2000).
[e] Lifetime exposure to ambient levels of 1,3-D in a high-use county (Merced), was estimated for various residency times and locations using computer modeling of air concentration and residency mobility data. The estimates for STAC and SAC were generated using air monitoring data obtained in Merced County (Rotondaro and Van Wesenbeeck, 2012a). AAC was estimated by simulation of the ambient air concentrations in Merced County (Rotondaro and Van Wesenbeeck, 2012a).

1. Handler and Reentry Worker Exposure Estimates

The registrants measured the 1,3-D breathing-zone air concentrations of applicators and loaders working with a tractor-mounted shank injection apparatus. The breathing-zone 1,3-D air concentrations of workers reentering the field after fumigation were also measured. These air monitoring studies were conducted in Washington, Arizona, and North Carolina. Following the fumigations, the fields were left uncovered (i.e., no tarp-seal). The injection depths ranged from 10 - 18 inches, and the application rates were 114, 165, and 237 lbs of active ingredient (AI)/acre (Houtman, 1993). Breathing-zone air concentrations were directly measured or derived from biomonitoring data (urinary metabolites). The directly measured air concentration data were used for estimating exposure values in this EAD. Two types of air-monitoring samples were generated, activity-specific samples with sampling periods ≤ 46 minutes, and samples with longer sampling periods of approximately 4 hours. The activity-specific samples were taken just during activities with high exposure potential, while the 4-hour samples included both high-exposure potential and other work-related activities conducted during the experiments. Being more representative of exposures occurring over the course of the full workday, the 4-hour TWA samples were the only samples utilized to assess the 8-hr TWA exposures in this EAD. These measured air concentrations were corrected for recovery, adjusted to the maximum legal application rate of 332 lbs of 1,3-D/acre, and, if applicable, adjusted for the use of respiratory protection. The STAC was made equal to the 95th %-ile of the natural logarithm values of these adjusted air concentrations (Frank, 2009). Alternatively, the SAC, AAC and LAC, are based upon the mean of the measured air concentrations which have been corrected for recovery, adjusted to the appropriate seasonal application rate and, when applicable, adjusted for the use of respiratory protection. As stated previously, the seasonal application rates and use seasons were derived from the latest 5 years (2010-14) of AGRIAN PUR data (DAS, 2011; DAS, 2012; DAS, 2013; DAS, 2014; DAS, 2015a).

a. Applicator (shallow shank without tarp)

Short-term air concentration

The registrant study utilized to estimate exposure contained applicator/driver 1,3-D breathing-zone air monitoring data for applications utilizing a tractor equipped with a shallow shank injection apparatus. The treated ground was not sealed with a tarp. The breathing-zone samples were obtained under three different conditions:

1. The tractor cab windows were open and injection shanks were fitted with spillage controls.
2. The tractor cab windows were closed and the cab was equipped with a carbon filtration unit. In addition, injection shanks were fitted with spillage controls.
3. The tractor cab windows were open and spillage controls were not fitted to the injection shanks.

The air monitoring data obtained under the first set of conditions was used for estimating exposure. While spillage controls are required when applying via shank injection in CA, the use of a closed-cab tractor is not. As mentioned earlier, the breathing-zone samples were obtained at three sites. At each site, a 4-hr TWA sample was obtained from each of 5 separate applications. A level equivalent to the 95^{th} %-ile of the natural logarithm of these air concentration data is not anticipated to occur on a daily basis. However, air concentrations this high may occur on a given workday (Frank, 2009). The calculated air concentration was adjusted to the allowed maximum application rate (i.e., 332 lbs AI/acre) allowed under CA permit conditions. According to the product labels, handlers must wear respiratory protection (half-face respirator). The assigned protection factor (APF) for this PPE is 10 as stated in the California Code of Regulations (CCR), Title 8, Section 5144 (CCR 8 5144) and by OSHA (OSHA, 2009). A respirator with an APF of 10 removes 90% of the air contaminant from the breathing-zone (Beauvais, 2011). Hence, the calculated air concentration was multiplied by 0.1 to adjust for the use of a half-face respirator to get 0.27 ppm (Table IV.8).

Seasonal air concentration

As mentioned earlier, the estimated use seasons and seasonal application rates for handlers were generated from the latest 5 years of the AGRIAN® PUR database records for 1,3-D. These estimates were made for the company applying the greatest amount of AI in the highest use county using the shallow shank method. As previously stated, the data for applications conducted with a tarp were combined with those for applications conducted without the use of a tarp. For the shallow shank application method, the use season and application rate are 3 months, with 85 application days within this season, and 154 lbs/acre. To estimate the SAC, the measured air concentrations at each of the 3 sites were adjusted to this seasonal application rate and for the use of a half-face respirator. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.032 ppm. Hence, the applicator using the shallow shank without the use of a tarp is anticipated to be exposed to a daily (8-hr TWA) air concentration of 0.032 ppm 1,3-D for 85 days during the use season (Table IV.8).

Annual air concentration

The total number of shallow shank application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 115. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.0096 ppm (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.0054 ppm (Table IV.8).

Table IV. 8 Occupational Exposure Estimates (ppm)

Exposure Scenario [a]	STAC [b]	SAC [c]	AAC [d]	LAC [e]
applicator (shallow shank w/o tarp)	0.27	0.032	0.0096	0.0054
applicator (shallow shank w/ tarp)	0.85	0.10	0.032	0.017
applicator (deep shank w/o tarp)	0.27	0.068	0.042	0.023
applicator (deep shank w/ tarp)	0.85	0.22	0.14	0.072
applicator (drip w/o tarp)	0.28	0.039	0.013	0.0070
applicator (drip w/ tarp)	0.23	0.018	0.0060	0.0032
applicator (injection auger)	1.2	n/a	n/a	n/a
loader (shallow shank)	0.70	0.062	0.019	0.0100
loader (deep shank)	0.70	0.13	0.082	0.044
tarp remover (shallow shank)	33	3.9	1.2	0.66
tarp remover (deep shank)	33	8.3	5.2	2.8
tarp remover (drip)	33	2.6	0.85	0.46
reentry worker (shallow shank)	0.037	0.015	0.0064	0.0034
reentry worker (deep shank)	0.037	0.032	0.024	0.013
reentry worker (drip)	0.037	0.010	0.0044	0.0024
occupational bystander (shallow shank w/o tarp)	2.0	0.0012	0.00062	0.0033
occupational bystander (deep shank w/o tarp)	0.6	0.0012	0.00062	0.0033
occupational bystander (drip w/ tarp)	1.1	0.0012	0.00062	0.0033

[a] The handler, with the exception of the tarp remover, exposure estimates incorporated the protection factor for a half-face respirator (90%), while the reentry worker and occupational bystander exposure estimates did not incorporate this protection factor.

[b] STAC: Short-Term Air Concentration: The estimated 8-hr TWA 1,3-D breathing-zone air concentration of the worker. This estimate consists of the 95th %-ile of the air concentration data, corrected for recovery, adjusted to the maximum application rate (332 lbs 1,3-D/acre), and, if applicable, for the use of a respirator.

[c] SAC: Seasonal Air Concentration: This estimate is the daily 8-hr TWA 1,3-D breathing-zone air concentration that the worker is anticipated to breathe over the course of the estimated use season . The estimate consists of the mean value of the air concentration data, corrected for recovery, adjusted to the appropriate seasonal application rate, and, if applicable, for the use of a respirator.

[d] AAC: Annual Air Concentration: This estimate represents the breathing-zone air concentration of the worker amortized over the full year. It is equal to the SAC multiplied by the ratio of the annual number of application days to the number of days in the year (i.e., 365).

[e] LAC: Lifetime Air Concentration: This estimate represents the working environment breathing-zone air concentration of the worker amortized over a lifetime. The LAC is equal to the AAC multiplied by the assumed number of years worked over a lifetime (i.e., 40 years), divided by the assumed lifespan (i.e., 75 years).

The surrogate data approach was used to estimate exposure for four types of applicators. These scenarios are the applicator using shallow shank with the use of a tarp, the applicator using deep shank with and without the use of a tarp, the applicator using the drip method with and without the use of a tarp, and the applicator utilizing the injection auger method. Another handler scenario assessed using surrogate data is the tarp remover. These surrogate data were obtained from two chloropicrin air monitoring studies reviewed previously by DPR staff, (Beauvais, 2010), and subsequently used for the chloropicrin exposure assessment document (Lewis, 2012). These studies were conducted by the Chloropicrin Manufacturers Task Force which measured breathing-zone air concentrations for various exposure scenarios including handlers conducting shank, drip, and injection auger fumigation, and tarp removal (Beard, 1996) (Rotondaro, 2004). However, the data collected at the Arizona site in the Beard et al. study were not used to generate the chloropicrin ratios. The applications done at this site did not meet the good agricultural practices requirement on the federal label (Barry, 2014).

The approach used to derive the 1,3-D breathing-zone air concentrations for the aforementioned scenarios consisted of adjusting the measured 1,3-D breathing-zone air concentrations for the applicator using a shallow shank (broadcast and without the use of a tarp), with a ratio of the surrogate data. The ratio consisted of the 95th %-ile of the measured chloropicrin breathing-zone air concentrations for the scenario of interest for 1,3-D exposure assessment over the 95th %-ile of the measured chloropicrin breathing-zone air concentrations for the applicator using shallow shank (broadcast and without the use of a tarp). An example, using the tarp remover scenario, of this approach which shows the assumed relationship between the chloropicrin (CP) and 1,3-dichloropropene air concentrations (1,3-D) is presented below:

$$\frac{\text{TARP REMOVER (CP)}}{\text{S.SHANK APPLICATOR W/O TARP (CP)}} \quad \alpha \quad \frac{\text{TARP REMOVER (1,3-D)}}{\text{S.SHANK APPLICATOR W/O TARP (1,3-D)}}$$

Based upon this assumed relationship, the 1,3-D breathing-zone air concentration for the tarp remover is derived from the measured 1,3-D breathing-zone air concentration for the shallow

shank without tarp applicator using the chloropicrin air concentration ratio:

$$\frac{\text{TARP REMOVER (CP)}}{\text{S.SHANK APPL. W/O TARP (CP)}} \times \text{S.SHANK APPL. W/O TARP (1,3-D)} = \text{TARP REMOVER (1,3-D)}$$

The chloropicrin air concentrations utilized in the ratio were corrected for recovery and adjusted to the same maximum application rate. The product of the multiplication between the measured 1,3-D breathing-zone air concentrations for the applicator using a shallow shank (broadcast and without the use of a tarp) and the ratio is the derived 1,3-D breathing-zone air concentration for the scenario of interest.

The surrogate ratio approach using chloropicrin is a reasonable first approximation of the 1,3-D air worker breathing zone air concentrations. Chloropicrin and 1,3-D do differ in their physical and chemical properties, and those differences produce differing patterns in mass loss following the application. However, both chloropicrin and 1,3-D tend to show small flux immediately following the application. For the majority of applications the maximum flux for both chloropicrin (Barry, 2014) and 1,3-D (Knuteson *et al.*, 1992b; Knuteson *et al.*, 1992a; Knuteson *et al.*, 1995; Gillis and Dowling, 1998; Knuteson and Dolder, 2000; Van Wesenbeeck and Phillips, 2000) occur 6 or more hours following application. In some studies the maximum flux occurs 24 hours or more following the application. During the application process the magnitude of flux will more likely be dominated by the application method itself. Application methods are reasonably standard between fumigants. The similarly small initial flux for most chloropicrin and 1,3-D applications supports this assumption and by extension, also supports the surrogate ratio approach.

The applicability of the chloropicrin tarp remover surrogate ratio depends upon the permeability of the tarps used and the soil degradation rate of chloropicrin relative to 1,3-D. USEPA conducted analysis of laboratory measured tarp permeability for various tarp types (Sarkar, 2010). Those results indicate that for 25 tarps, chloropicrin permeability is lower than 1,3-D permeability. Chloropicrin and 1,3-D differ in their soil degradation rate, 3.5 days versus 7 days, respectively (Johnson and Spurlock, 2012). However, modeling analysis conducted with the HYDRUS soil physics model indicates that chloropicrin and 1,3-D 6-hr flux were within a factor of 2 for both a 5 day and a 10 day tarp cutting interval (Johnson and Spurlock, 2012). Thus, for tarp remover exposure scenarios chloropicrin is a reasonable 1,3-D surrogate.

b. Applicator (shallow shank with tarp)

Short-Term air concentration

The breathing-zone air concentration for the applicator utilizing a shallow shank and a tarp for sealing the treated soil was not measured by the registrant. Since no direct-measurement data were available, breathing-zone data measured in the aforementioned chloropicrin studies were utilized as surrogate data for this exposure scenario. As described earlier, a ratio was generated

from the chloropicrin air concentration data. The 95th %-ile of the natural logarithms of the breathing-zone air concentrations measured for the applicator injecting chloropicrin using a shallow shank and then sealing the treated soil with a tarp was adjusted for recovery and the allowed maximum application rate to get 1948 µg/m^3. This value was divided by the corresponding value for the applicator injecting chloropicrin using a shallow shank without the use of a tarp (i.e., 613 µg/m^3) to obtain the ratio. This value was then multiplied with the measured air concentrations obtained, with spill controls in place, from the three study sites. Ideally, the 1,3-D air monitoring data obtained without the use of spillage controls should be used since they correspond with the assumed conditions of the chloropicrin study (i.e., no spillage controls). However, since this scenario represents the handler using shank application and spillage controls are required in CA, the 1,3-D monitoring data obtained with spillage controls in place were utilized to estimate exposure. The derived air concentrations were adjusted to the maximum application rate allowed under CA permit conditions, and for the use of a half-face respirator. The 95th %-ile of these adjusted air concentrations is 0.85 ppm (Table IV.8).

Seasonal air concentration

As previously stated, for the shallow shank application method, the use season and application rate are 3 months, with 85 application days within this season, and 154 lbs/acre. To estimate the SAC, the measured air concentrations at each of the 3 sites were adjusted to this seasonal application rate, the use of a half-face respirator, and multiplied by the previously described ratio. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.10 ppm. Hence, the applicator using the shallow shank without the use of a tarp is anticipated to be exposed to a daily (8-hr TWA) air concentration of 0.10 ppm 1,3-D for 85 days during the use season (Table IV.8).

Annual air concentration

As stated earlier, the total number of shallow shank application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 115. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.032 ppm (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.017 ppm (Table IV.8).

c. Applicator (deep shank without tarp)

As stated earlier, the AGRIAN PUR data from 2010-14 showed that the bulk of 1,3-D applications were done using the deep shank method. As a result, this exposure scenario, with and without the use of tarp, was added to the current draft. Due to a lack of data, the STAC generated for the applicator using shallow shank without a tarp was used as the STAC for the applicator using deep shank without a tarp. However, the estimated seasonal application rate and

use information for the applicator using deep shank differ from those of the applicator using shallow shank. Hence, the SAC, AAC, and, as a result, LAC values differ between these two scenarios.

Short-Term air concentration
Due to a lack of data, the STAC generated for the applicator using the shallow shank method without the use of a tarp is also the STAC for the applicator using the deep shank method without the use of a tarp (Table IV.8).

Seasonal air concentration
For the deep shank application method, the use season and application rate are 8 months, with 193 application days within this season, and 327 lbs/acre. To estimate the SAC, the measured air concentrations for the applicator using shallow shank without the use of a tarp at each of the 3 sites were adjusted to this seasonal application rate and for the use of a half-face respirator. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.068 ppm. Hence, the applicator using the deep shank without the use of a tarp is anticipated to be exposed to a daily (8-hr TWA) air concentration of 0.068 ppm 1,3-D for 193 days during the use season (Table IV.8).

Annual air concentration
The total number of deep shank application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 228. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.042 ppm (Table IV.8).

Lifetime air concentration
The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.023 ppm (Table IV.8).

d. Applicator (deep shank with tarp)

Short-Term air concentration
Due to a lack of data, the STAC generated for the applicator using the shallow shank method with the use of a tarp is also the STAC for the applicator using the deep shank method with the use of a tarp (Table IV.8).

Seasonal air concentration
For the deep shank application method, the use season and application rate are 8 months, with 193 application days within this season, and 327 lbs/acre. To estimate the SAC, the air concentrations for the shallow shank applicator using a tarp, at each of the 3 sites, were adjusted to this seasonal application rate and for the use of a half-face respirator. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.22 ppm. Hence, the applicator using the deep shank with the use of a tarp is anticipated to be

exposed to a daily (8-hr TWA) air concentration of 0.22 ppm 1,3-D for 193 days during the use season (Table IV.8).

Annual air concentration

The total number of deep shank application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 228. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.14 ppm (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.072 ppm (Table IV.8).

c. Applicator (drip without tarp)

Short-Term Air Concentration

The breathing-zone air concentrations for the applicator utilizing drip chemigation without the use of a tarp for sealing the treated soil was not measured by the registrant. Since no direct-measurement data were available, breathing-zone data measured in a chloropicrin study were utilized as surrogate data for this exposure scenario. As described earlier, a ratio was generated from the chloropicrin air concentration data. The 95th %-ile of the natural logarithms of the breathing-zone air concentrations measured for the handler applying chloropicrin using a drip chemigation without the use of a tarp (i.e., 305 µg/m^3 at an application rate of 300 lbs AI/acre) was adjusted for recovery and to the allowed maximum application rate for shank (i.e., 500 lbs AI/acre), to get an adjusted value of 508 µg/m^3. This value was then divided by the corresponding value for the applicator injecting chloropicrin using a shallow shank without the use of a tarp (i.e., 613 µg/m^3) to obtain the ratio. This value was then multiplied with the measured air concentrations obtained, without spill controls in place, from the three study sites. Of the registered products containing 1,3-D, only one allows for the omission of a soil-sealing tarp during fumigation. This product is a mixture of 1,3-D and chloropicrin and is called Pic-Clor 60 EC, with 1,3-D comprising 37.1% of the product. The maximum product label application rate for this product translates into 187 lbs 1,3-D/acre. According to the CA permit conditions for 1,3-D, since this maximum product label rate is less than that allowed by CA permit conditions (i.e., 332 lbs/acre), the product label rate becomes the highest legal rate (CDPR, 2015c). The aforementioned derived air concentrations were adjusted to this maximum product label rate for 1,3-D and for the use of a half-face respirator. The 95th %-ile of these adjusted air concentrations is 0.28 ppm (Table IV.8).

Seasonal Air Concentration

For the drip application method (with and without the use of a tarp), the use season and application rate are 3 months, with 73 application days within this season, and 101 lbs/acre. To estimate the SAC, the air concentrations for the shallow shank applicator, without the use of a

tarp, at each of the 3 sites, were adjusted to this seasonal application rate, for the use of a half-face respirator, and multiplied by the previously described ratio. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.039 ppm. Hence, the applicator using the drip method without the use of a tarp is anticipated to be exposed to a daily (8-hr TWA) air concentration of 0.039 ppm 1,3-D for 73 days during the use season (Table IV.8).

Annual air concentration

The total number of drip application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 121. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.013 ppm (Table IV.8).

Lifetime Air Concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.0070 ppm (Table IV.8).

d. Applicator (drip with tarp)

Short-Term air concentration

The breathing-zone air concentrations for the applicator utilizing drip chemigation with the use of a tarp for sealing the treated soil was not measured by the registrant. Since no direct-measurement data were available, breathing-zone data measured in a chloropicrin study were utilized as surrogate data for this exposure scenario. As described earlier, a ratio was generated from the chloropicrin air concentration data. The 95th %-ile of the natural logarithms of the breathing-zone air concentrations measured for the handler applying chloropicrin using drip chemigation with the use of a tarp (i.e., 141 µg/m^3 at an application rate of 300 lbs AI/acre) was adjusted for recovery and to the allowed maximum application rate for shank (i.e., 500 lbs AI/acre), to get an adjusted value of 235 µg/m^3. This value was then divided by the corresponding value for the applicator injecting chloropicrin using a shallow shank without the use of a tarp (i.e., 613 µg/m^3) to obtain the ratio. The measured 1,3-D air concentrations for the applicator using shallow shank without tarp and spillage controls were multiplied by this adjustment factor to get the derived air concentrations for the handler applying 1,3-D using drip chemigation with the use of a tarp for sealing the soil. Of the three registered drip application products containing 1,3-D, two of them require the use of a tarp during application and fumigation for sealing the treated soil. One of the products has a maximum application rate of 392 lbs 1,3-D/acre which exceeds the maximum application rate of 332 lbs 1,3-D/acre allowed under CA permit conditions. Hence, the maximum CA 1,3-D permit condition application rate of 332 lbs AI/acre was used to estimate exposure (CDPR, 2015c). The aforementioned derived air concentrations were adjusted to this maximum application rate for 1,3-D and for the use of a half-face respirator. The 95th %-ile of these adjusted air concentrations is 0.23 ppm (Table IV.8).

Seasonal air concentration

For the drip application method (with and without the use of a tarp), the use season and application rate are 3 months, with 73 application days within this season, and 101 lbs/acre. To estimate the SAC, the air concentrations for the shallow shank applicator, without the use of a tarp, at each of the 3 sites, were adjusted to this seasonal application rate, for the use of a half-face respirator, and multiplied by the previously described ratio. The mean of these adjusted values was taken for each site. The mean of the three means is equal to the SAC value of 0.018 ppm. Hence, the applicator using the drip method with the use of a tarp is anticipated to be exposed to a daily (8-hr TWA) air concentration of 0.018 ppm 1,3-D for 73 days during the use season (Table IV.8).

Annual air concentration

The total number of drip application days, with and without the use of a tarp, for the company applying the most fumigant in the highest use county is 121. To obtain the AAC, this value was divided by the number of days in the entire year (i.e., 365 days) and multiplied by the SAC to get 0.0060 ppm (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.0032 ppm (Table IV.8).

e. Applicator (injection auger)

Short-Term air concentration

The breathing-zone air concentrations for the applicator utilizing injection auger application were not measured by the registrant. Since no direct-measurement data were available, breathing-zone data measured in the aforementioned chloropicrin study were utilized as surrogate data for this exposure scenario. As described earlier, a ratio was generated from the chloropicrin air concentration data. The 95^{th} %-ile of the natural logarithms of the breathing-zone air concentrations measured for the handler applying chloropicrin using an injection auger was adjusted for recovery and to the allowed maximum application rate (i.e., 500 lbs AI/acre), to get a value of 751 $\mu g/m^3$ (Beauvais, 2010). This value was then divided by the corresponding value for the applicator injecting chloropicrin using a shallow shank without the use of a tarp (i.e., 613 $\mu g/m^3$) to obtain the ratio.

Four of the products containing 1,3-D (i.e., Telone C-17, Telone C-35, Telone II, and Tri-Cal Trilone II), allow for injection auger application of 1,3-D to tree replant sites. The amount of product to be applied per replant site as specified on the product labels ranges from 24 to 33 ounces while the concentration of AI in the four products ranges from 7.1 to 9.85 lbs AI/gal. The maximum allowable application rate of 332 lbs 1,3-D/acre as stated on product labels and the CA permit conditions does not apply to the injection auger application method. Hence, the maximum application rate for this method was derived by estimating the amount of time required to treat a

given tree replant site and the number tree replant sites that an applicator could potentially treat in an 8-hr workday. This information was obtained from the aforementioned chloropicrin hand-wand applicator exposure study where the applicator treated tree replant sites of the same dimensions as those specified on the 1,3-D product labels (i.e, 10 x 10 foot squares) (Beauvais, 2010). In the study, the applicators managed to treat from 48 to 96 replant sites over 2.0 to 3.1 hours. These values are equivalent to 24 to 31 tree replant sites/hr. Over an 8-hr work period, 31 tree replant sites/hr is equivalent to 248 replant sites. An acre could potentially contain 436 ten x ten foot replant sites. Hence, one acre could contain the estimated 248 treated replant sites (248 replant sites/acre). Multiplying the product label specified number of ounces to be applied to each replant site with the conversion factor (1 gal/128 fluid ounces), the product 1,3-D concentration (lbs AI/gal), and the number of tree replant sites treated over an 8-hr period, generates the derived maximum application rate for hand-wand application in lbs of 1,3-D/acre (Table IV.9).

Table IV. 9 Maximum 1,3-D Hand-Wand Injection Rates for Tree Replant Site Fumigation

Product	Product Label Application Rate (fl oz/10'x10' Replant Site) [a]	Product 1,3-D Concentration (lbs AI/gal)	Max Appl Rate (lbs 1,3-D/acre) [b]
Telone C-17	31	8.6	517
Telone C-35	33	7.1	454
Telone II	24	9.85	458
Tri-Cal Trilone II	24	9.85	458

[a] fl oz = fluid ounces

[b] Maximum application rate in pounds 1,3-D per acre = Max Appl Rate (*lbs 1,3-D/acre*) = (fl oz per 10ft x 10ft replant site) x (1gal/128 fl oz) x (lbs 1,3-D/gal) x (248 replant sites/acre)

To estimate the short-term breathing air concentration for the hand-wand applicator, the measured 1,3-D air concentrations for the applicator using shallow shank without a tarp and spillage controls, corrected for recovery, and adjusted to the highest of the estimated application rates in Table IV.7 (i.e., 517 lbs 1,3-D/acre), were multiplied by the aforementioned surrogate ratio. These air concentrations were then adjusted for the use of a half-face respirator. The 95th %-ile of these adjusted air concentrations is 1.2 ppm (Table IV.8).

Seasonal, annual, and lifetime air concentrations

The applications listed in the AGRIAN PUR records from 2010-14 are described as being conducted via shallow shank, deep shank, drip, and "unknown/other". As mentioned earlier, the number of pounds applied from 2010-14 for the unknown/other category is 725,718. Due to the relatively low number of pounds that can be applied via the injection auger method and the likely sporadic use of this method for tree and vine replacement, the bulk of the pounds applied for unknown/other is likely due to shank or drip. Hence, seasonal, annual, and lifetime exposures for the applicator using the injection auger method are not anticipated.

f. Loader

Short-Term air concentration

The registrant study utilized to estimate exposure contained loader 1,3-D breathing-zone air monitoring data for applications utilizing a tractor equipped with a shank injection apparatus. The treated ground was not sealed with a tarp. The breathing-zone samples were obtained under three different conditions:

1. Dry disconnect fittings were used
2. Dry disconnect fittings and a vapor recovery system were used
3. Dry disconnect fittings and a vapor recovery system were not used

The air monitoring data obtained under the first set of conditions was used for estimating exposure (i.e., dry disconnect fittings). While dry disconnects are required when applying 1,3-D in CA, the use of a vapor recovery system is not. As mentioned earlier, the breathing-zone samples were obtained at three sites. At each site, a 4-hr TWA sample was obtained from each of 5 separate applications. As mentioned earlier, the STAC is equal to the 95^{th} %-ile of the natural logarithm of the air concentration data or 0.70 ppm (Table IV.8). The air monitoring data were obtained for the loader involved in shallow shank injection. However, due to the lack of data, they were used as surrogate data for the loader involved in deep shank injection.

Seasonal air concentration

As mentioned earlier, two 1,3-D use seasons were derived from the AGRIAN PUR database for the shank injection method. The use season for shallow shank injection is 3 months (August-October), while the season for deep shank injection is 8 months (January-March and August-December). The numbers of application days in these two seasons are 85 and 193 for the shallow shank and deep shank methods, respectively. The estimated seasonal application rates for shallow and deep shank are 154 and 327 lbs/acre, respectively. To estimate the SAC for each method, the measured air concentrations at each of the 3 sites were adjusted to the appropriate seasonal application rate and for the use of a half-face respirator. The mean of these adjusted values was taken for each site. For the shallow shank loader, the mean of the three means is equal to the SAC value of 0.062 ppm. For the deep shank loader, this value is 0.13 ppm. Hence, the shallow shank loader is anticipated to be exposed to a daily (8-hr TWA) 1,3-D air concentration of 0.062 ppm for 85 days during the use season. The deep shank loader is anticipated to be exposed to a daily (8-hr TWA) 1,3-D air concentration of 0.13 ppm for 193 days during the use season (Table IV.8).

Annual air concentration

The numbers of application days throughout the year for the shallow shank and deep shank methods are 115 and 228. Dividing each number by 365 days and multiplying each ratio with the respective SAC values generates AAC values of 0.019 and 0.082 ppm for the shallow shank and deep shank loaders, respectively (Table IV.8).

Lifetime Air Concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC values for the shallow shank and deep shank loaders are 0.010 and 0.044 ppm, respectively (Table IV.8).

g. Tarp Remover

Short-Term air concentration

The breathing-zone air concentrations for the worker removing tarps were not measured by the registrant. Since no direct-measurement data were available, breathing-zone data measured in the aforementioned chloropicrin study were utilized as surrogate data for this exposure scenario. As described earlier, a ratio was generated from the chloropicrin air concentration data. The 95th %-ile of the natural logarithms of the breathing-zone air concentrations measured for the handler removing the tarp from a fumigated field was adjusted for recovery and to the allowed maximum application rate (i.e., 500 lbs AI/acre), to get a value of 3319 µg/m^3. This value was then divided by the corresponding value for the applicator injecting chloropicrin using a shallow shank without the use of a tarp (i.e., 613 µg/m^3) to obtain the ratio. The measured 1,3-D air concentrations for the applicator using shallow shank without tarp and spillage controls, corrected for recovery and adjusted to the maximum application rate, were multiplied by this adjustment factor to get the derived air concentrations for the tarp remover.

Respiratory protection was not incorporated into the exposure estimate for the tarp remover scenario. Five of the labels (Telone II, Pic-Clor 60 EC, Trilone II, Telone EC, and Inline) for active products on the DPR product label database for 1,3-D allow for exposures without respiratory protection. As stated in all five of these product labels, from 1-5 days following application, a half-face respirator is required for the handler (e.g., tarp remover), entering the treated area. However, for three of these product labels (Telone II, Tri-Cal Trilone II, and Telone EC), there's no information on respirator requirements for handlers entering the treated area after the 5-day fumigation period. The other two labels (Pic-Clor 60 EC and Inline), state that the handler entering the treated area 5 days or more after the application doesn't have to don respiratory protection unless irritation of the eyes and nose, presumably due to the chloropicrin in the product mixture, occurs. Moreover, the chloropicrin tarp remover breathing-zone air monitoring data used to derive the 1,3-D air concentrations was collected 7 days post-application. Due to these issues and the fact that CA permit conditions for 1,3-D do not address this particular scenario, a respiratory protection factor for the half-face respirator was not incorporated into the exposure estimate calculations. The 95th %-ile of the natural logarithm of these adjusted air concentrations is 33 ppm (Table IV.8). Due to a lack of data, this value was used to represent the STAC for the handler removing tarps from fields treated with the shallow shank, deep shank, or drip methods.

Seasonal air concentration

The tarp remover could potentially be removing tarps from fields treated using shallow shank, deep shank, or drip fumigation. In addition, use seasons for each method were generated. Hence,

a SAC value was generated for the tarp remover scenario for each application method. Using the aforementioned use seasons and application rates for these application methods, and the mean of the derived tarp remover breathing-zone air concentrations, the SAC values for the handler removing tarps from fields that have been treated via shallow shank, deep shank, or drip are 3.9, 8.3, and 2.6 ppm, respectively. Hence, the tarp remover is anticipated to be exposed to an air concentration of 3.9 ppm for 8-hr TWA for 85 days during the use season for shallow shank, 8.3 ppm for 193 days during the use season for deep shank, and 2.6 ppm for 73 days during the use season for drip (Table IV.8).

Annual air concentration

The numbers of application days throughout the year for the shallow shank, deep shank, and drip methods are 115, 228, and 121. Dividing each number by 365 days and multiplying each ratio with the respective SAC values generates AAC values of 1.2, 5.2, and 0.85 ppm for handlers removing tarps from fields treated using the shallow shank, deep shank, and drip methods, respectively (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC values for the handler removing tarps from fields treated using the shallow shank, deep shank, and drip application methods are 0.66, 2.8, and 0.46 ppm, respectively (Table IV.8).

h. Reentry worker

Short-Term air concentration

The registrant study utilized to estimate exposure contained reentry worker 1,3-D breathing-zone air monitoring data for shank injections which were conducted without the use of a soil-sealing tarp. The breathing-zone samples were obtained for reentry workers at three different sites conducting three different activities: center pivot maintenance and winterization about 3.8 days following fumigation, rock removal at approximately 2.7 days post-fumigation, and bed shaping at 3 to 24 hours post-fumigation. The CA restricted entry interval (REI) for 1,3-D containing products is 7 days. The reentry worker air monitoring study conducted 3.8 days post-fumigation is closest to this period and was used to estimate reentry worker exposure. Four-hour TWA samples were obtained from 5 reentry workers who were maintaining and winterizing the center pivots. The measured air concentrations, adjusted to the maximum application rate, are 0.032, 0.034, 0.031, 0.028, and 0.036 ppm. As mentioned earlier, for the STAC, the 95^{th} %-ile of the natural logarithm of the data is used to calculate exposure. The 95^{th} %-ile value of the natural logarithm of these five values is 0.037 ppm. Due to a lack of data, this STAC value was used as a surrogate STAC for the worker reentering a field treated using the deep shank and drip methods (Table IV.8).

Seasonal air concentration

The reentry worker could potentially reenter fields treated using shallow shank, deep shank, or drip fumigation. In addition, use seasons for each method were generated. Hence, a SAC value was generated for the reentry worker scenario for each application method. Using the aforementioned use seasons and application rates for these application methods, and the mean of the means of the breathing-zone air concentrations, the SAC values for the worker reentering fields that have been treated via shallow shank, deep shank, or drip are 0.015, 0.032, and 0.010 ppm, respectively. Hence, the reentry worker is anticipated to be exposed to an air concentration of 0.015 ppm for 8-hr TWA for 108 days during the use season for shallow shank, 0.032 ppm for 203 days during the use season for deep shank, and 0.010 ppm for 82 days during the use season for drip (Table IV.8).

Annual air concentration

The numbers of application days throughout the year for the shallow shank, deep shank, and drip methods are 159, 283, and 163. Dividing each number by 365 days and multiplying each ratio with the respective SAC values generates AAC values of 0.0064, 0.024, and 0.0044 ppm for workers reentering fields treated using the shallow shank, deep shank, and drip methods, respectively (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC values for the worker reentering fields treated using the shallow shank, deep shank, and drip application methods are 0.0034, 0.013, and 0.0024 ppm, respectively (Table IV.8).

2. Occupational Bystander Exposure Estimates

To generate the 8-hr TWA occupational bystander exposure estimates, 1,3-D air concentrations were simulated using the Industrial Source Complex Short-Term version 3 (ISCST3) program. The air concentrations associated with the maximum 8-hr flux during daylight hours were simulated for estimating exposure. In addition, the simulations incorporated the maximum application rate allowed on the permit conditions for shank application. Simulated 1,3-D air concentrations were generated for shallow shank (< 18 inches injection depth) and deep shank (≥ 18 inches injection depth) injection, both without the use of soil-sealing tarps (Johnson, 2002). The acreages used for the simulations are 80 and 40 acres for the shank and drip application methods, respectively. For drip application, the application rate of 252 lbs/acre was utilized. The two drip fumigation air monitoring studies used for the simulation had tarp seals in place (Johnson, 2002; Johnson, 2009a). Of the four registered drip application products containing 1,3-D, two of them require the use of a tarp during application and fumigation for sealing the treated soil. One of these two products has a maximum application rate of 392 lbs 1,3-D/acre which exceeds the maximum application rate of 332 lbs 1,3-D/acre allowed under CA permit conditions (CDPR, 2015c). Hence, the simulated air concentration for drip chemigation was adjusted to 332 lbs 1.3-D/acre for estimating exposure.

Short-Term air concentration

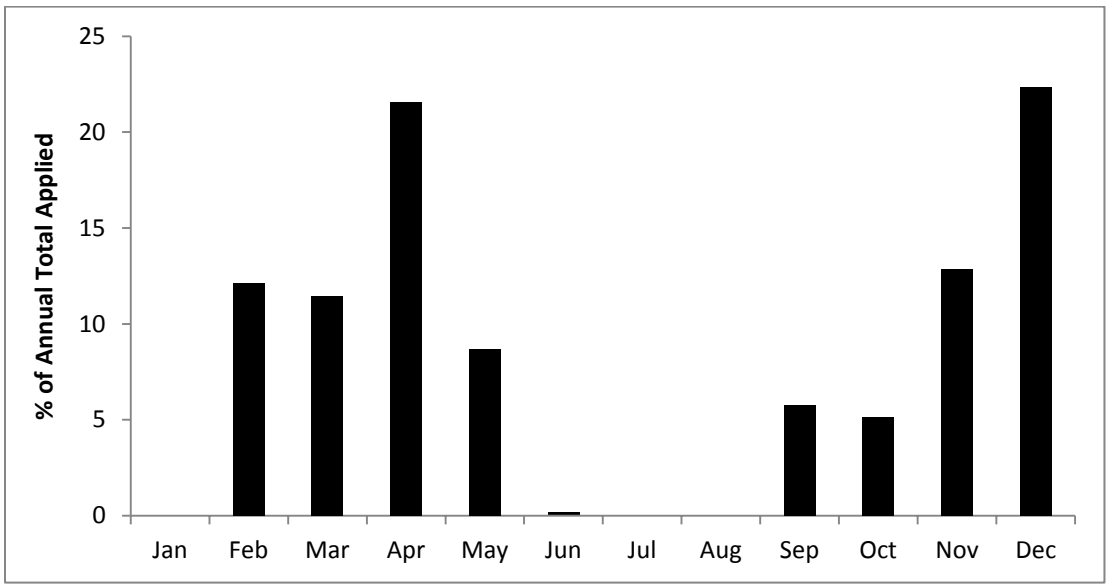
As indicated earlier, the occupational bystander exposure estimates were generated using simulated 1,3-D air concentrations. Since buffer-zone restrictions were not considered for the occupational bystander in the field, the simulated air concentrations located closest to field-edge (i.e., 3 meters or 9.97 feet), were used to estimate exposures. In addition, the occupational bystander is not required to wear respiratory protection so the corresponding protection factor was not incorporated into the exposure estimate calculation. The simulations were conducted for shallow shank, deep shank, and drip chemigation. The shank application studies used for modeling did not utilize a soil-sealing tarp (Johnson, 2002). The drip chemigation studies used for the simulation did use soil-sealing tarps (Johnson, 2002; Johnson, 2009a). The air concentrations were generated using a nominal flux of 100 $\mu g/m^2/s$ for all applications and all field sizes (Johnson, 2009a). This allowed for scaling the air concentrations to the maximal application rate (Barry, 2015b). The scaling was done by using the ratio 332/252=1.317. The treated field sizes for the simulations were 80 acres and 40 acres for shank, and drip fumigation, respectively. The 8-hr TWA air concentrations generated for shallow shank, deep shank, and drip chemigation are 2 ppm, 0.6 ppm, and 1.1 ppm, respectively (Table IV.8).

Although the occupational bystander is not anticipated to be adjacent to a field undergoing fumigation on a daily basis, long-term exposure to ambient levels of 1,3-D may occur throughout the use season and year. To assess these exposures, measured air concentration data obtained by the registrant in Merced County (one of the 5 highest use counties) from January through December 2011 were utilized. The annual number of pounds of AI applied in this county was 1.5 times the township cap set by DPR. The air sampling was conducted in square 9- township area within the county. Four of the townships have historically high use of 1,3-D while the 5 other townships, adjacent to the high-use townships, have historically low to moderate use of 1,3-D. The samplers were placed near the center of each township, referred to in the study as a "receptor". The sites consisted of a dairy, two chicken ranches, a cattle feedlot surrounded by orchards, wildlife preserves, and locations near orchards. The samplers were placed at a height of 1.5 meters above ground. Each sample was collected through charcoal tube at a flow rate of 1L/hr over a 72-hr period. The sample-containing tube was then collected and immediately replaced with a new charcoal tube to continue the sampling process. The sample tubes contained two sections with activated charcoal. Breakthrough of sample from the front section to the back section occurred in <1% of all of the sample tubes. In these cases, the levels of breakthrough detected were less than the LOQ. Field-fortification samples were conducted for the study to estimate sample recoveries. The overall average sample recoveries were 90% and 84% for cis- and trans-isomers of 1,3-D, respectively. Of the 9 receptors in the study, the samples collected from Receptor 5 had the highest mean air concentration of 1,3-D (Rotondaro and Van Wesenbeeck, 2012a).

Merced County exceeded the annual township cap by 1.5 times. However other counties showed higher annual total number of pounds. For comparison, in 2011, the annual total number of

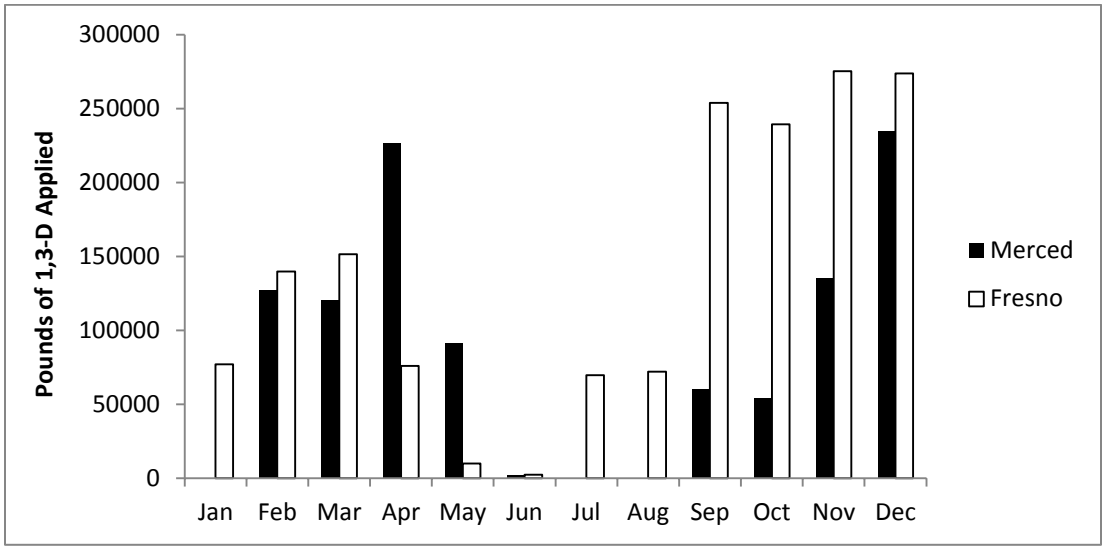
pounds of AI applied in Fresno County exceeded the annual total number of pounds of 1,3-D applied in Merced County. In 2011, 1,639,683 lbs of 1,3-D were applied in Fresno County while 1,050,205 lbs of 1,3-D were applied in Merced County. Since the annual total amount of 1,3-D applied in Merced County exceeded the annual township cap by 1.5 times, the entire year of 1,3-D air concentration samples in the county were not used. This annual rate of application may have generated exposure conditions higher than a typical seasonal exposure. Hence, this analysis used the Merced County air concentrations averaged over months grouped by season rather than air concentrations averaged over the entire year.. Merced had two use seasons in 2011 for a total use season of 8 months. One use season occurred from February through May while the other occurred from September through December. The amount of 1,3-D applied during each of these months was equal to or greater than 5% of the annual total 1,3-D applied in the county (Figure IV.1).

Figure IV.1. Monthly application of 1,3-D in Merced County in 2011



In 2011, from September through December the monthly number of pounds of 1,3-D applied in Fresno County exceeded the monthly totals for these months in Merced County (Figure IV.2).

Figure IV. 1 Monthly 1,3-D use in Fresno and Merced counties



Hence, even though the annual amount applied in Merced County exceeded the annual township cap, the total pounds applied over these individual months are less than the total pounds applied in Fresno County over the same months and, thus, were used to estimate the seasonal air concentration.

Seasonal air concentration

The SAC exposure estimate for the occupational bystander was calculated using the seasonal mean air concentration. The two use seasons in Merced County for 2011 consist of February-May and September through December. Of the two seasons, only the second season has monthly applied totals which are less than those of Fresno County for that year. Hence, the air concentrations measured during this use season were used to estimate exposure. For the months of September through December, the mean of the 1,3-D air concentrations measured in all 9 receptors in Merced County in 2011 is 0.0012 ppm (Table IV.8).

Annual air concentration

The AAC was estimated using the mean of air monitoring data from Merced County. This data was for the months of February through March, and September through December. The amounts of 1,3-D applied during each of these months were equal to or greater than 5% of the annual total. In addition, the monthly amounts applied were less than the amounts applied for the corresponding months in Fresno County in 2011. Hence, the SAC was multiplied by the ratio of 6 months/12 months to get an AAC of 0.00062 ppm (Table IV.8).

Lifetime air concentration

The LAC is obtained by multiplying the AAC with the number of years the handler is anticipated to work (i.e., 40) over the assumed lifetime of 75 years. The LAC is equal to 0.00033 ppm (Table IV.8).

3. Residential Bystander Exposure Estimates

The majority of 1,3-D product labels and CA permit conditions mandate a 100-foot buffer zone between the fumigated field and occupied structures. All but three labels (Telone EC, Telone II and TriCal Trilone II) also require that all non-handlers, including field workers, residents, pedestrians, and other bystanders, must be excluded from the buffer zone during the buffer zone period. All residential bystander exposures presented in this document are based on the assumption that a resident will spend 24 continuous hours either at 100 feet from a treated field, or in an area with elevated ambient air concentration of 1,3-D, or both. Short-term exposure is defined as exposure lasting from a day or less, and up to one week. Seasonal exposure is defined as a period of frequent exposure lasting more than a week but substantially less than a year, whether the exposure is constant or intermittent during the period. Annual exposure integrates all exposure periods during the year. Lifetime exposures integrate all exposure periods over several years (Beauvais, 2006; Beauvais, 2012).

Inhalation exposures of bystanders residing in close vicinity to the fumigated fields were calculated using the 1,3-D air concentrations based on ISCST3 modeling of the volatility loss (i.e., flux) at 100 ft. The fumigation methods considered in the modeling were: shallow shank, deep shank, and drip application. Tree and vine applications (based on all tree and all grape applications) constituted approximately 34% of the total use of 1,3-D during the period from 2008 to 2012 (Table II.6, 23.5% fruit/nut trees + 10.5% grapes). Because of the extensive 1,3-D use of tree and vine applications, separate calculations for the bystander exposure were performed.

a. Residential Bystander Exposures to Shank and Drip Fumigations

Short-Term air concentration

Twenty-four hour air concentrations were used for calculating short-term residential bystander inhalation exposures. The air concentrations at 100 ft downwind from shallow shank and deep shank applications were modeled for a field size of 80 acres at the maximum allowed application rate of 332 pounds per acre (Johnson, 2009a). The air concentrations at 100 ft downwind from drip applications were modeled for a field size of 40 acres and at an application rate of 252 pounds per acre, which is approximately the maximal application rate for strawberries on the InLine product label (Johnson, 2009a). The air concentrations were generated using a nominal flux of 100 $\mu g/m^2/s$ for all applications and all field sizes (Johnson, 2009a). This allowed for scaling the air concentrations to the maximal application rate (Barry, 2015b) . The scaling was done by using the ratio 332/252=1.317. The respective short-term air concentrations (STAC) calculated for shallow shank, deep shank and drip applications are 2500, 650 and 817 $\mu g/m^3$, or 0.5508, 0.1432 and 0.1800 ppm, respectively (Table IV.10).

Table IV. 10 Residential Bystander Exposure Estimates (ppm)

Exposure scenario	STAC	SAC	AAC	LAC
Exposure from nearby application site (edge of buffer zone), shallow shank application	0.5508 [a]	0.0173 [e]	N/D [h]	N/D [k]
Exposure from nearby application site (edge of buffer zone), deep shank application	0.1432 [a]	0.0135 [e]	N/D [h]	N/D [k]
Exposure from nearby application site (edge of buffer zone), drip application	0.1800 [b]	0.0050 [e]	N/D [h]	N/D [k]
Exposure from nearby application site (edge of buffer zone), tree and vine application	0.0918 [c]	N/D [f]	N/D [i]	N/A[l]
Exposure from ambient air	0.0813 [d]	0.0045 [g]	0.0002 [j]	Table IV.8[m]

[a] STAC: Short-Term Air Concentration: air concentrations are estimated at 100 feet downwind from the edge of a 1,3-D treated field, using short term air dispersion modeling, maximum application rate (332 lbs 1,3-D/acre), and field size of 80 acres (Johnson, 2009a).

[b] STAC: air concentrations are estimated at 100 feet downwind from the edge of the field, using short-term air dispersion modeling, maximum application rate (332 lbs 1,3-D/acre), and field size of 40 acres(Johnson, 2009a).

[c] STAC: air concentrations are estimated at 100 feet downwind from the edge of the field, using short-term air dispersion modeling and an application rate of 35 gal Telone II per acre (344 lbs 1,3-D/acre) (Powell, 2000).

[d] STAC: the highest 72-hour measured air concentration at Township # 5 recorded in Merced county between 01/03/2011 and 01/01/2012 (Rotondaro and Van Wesenbeeck, 2012a).

[e] SAC: Seasonal Air Concentration: air concentrations are estimated at 100 feet downwind from the edge of the field, using the two-week flux modeling as described in (Johnson, 2009b) but adjusted for median application rate and field size. The seasonal application rates were calculated as described in the Occupational Bystander Exposure section. The seasonal application rates were 153, 327 and 103 lb 1,3-D/acre for shallow shank, deep shank and drip application, respectively. The median field size for each application method was calculated from the DowAgrosciences PUR database for 1,3-D use California in 2010-2014 (Dow Agrosciences, 2011; Dow Agrosciences, 2012; DAS, 2014; Dow Agrosciences, 2014; Dow Agrosciences, 2015). The median field sizes were 20 acres, 15 acres, and 20 acres for shallow shank, deep shank and drip application, respectively.

[f] SAC: not determined.

[g] SAC: Seasonal Air Concentration: mean of air concentrations measured during the continuous use season of 4 months (September-December) in Merced County (Rotondaro and Van Wesenbeeck, 2012a).

[h] AAC: Annual Air Concentration; see text.

[i] AAC: not determined.

[j] AAC: median of 129,600 simulated annual ambient air concentration values of Township #5 by SOFEA-2 (SOFEA simulation as described in Section IV.B.3.d. Residential bystander exposure from ambient air (modeling).

[k] LAC: Lifetime Air Concentration; see text.

[l] LAC: not applicable because of the limited potential for life-time exposure.

[m] Multiple LAC values were generated in the following section for this exposure scenario using a computer modeling approach (Section IV.B.3.c, Table IV.8.

Seasonal air concentration

For consistency with previous fumigant exposure assessment (Beauvais, 2012) the seasonal air concentration estimate for residential bystanders was based on the two week 1,3-D generic air concentrations modeled using a nominal flux (100 µg/m^{2}sec) and field sizes as described in (Johnson, 2009b). We adjusted the 1,3-D generic air concentrations to reflect the seasonal application rate. The median field size for each application method was calculated from the PUR database provided by Dow AgroSciences for 1,3-D use in California in 2010-2014 (Dow

Agrosciences, 2011; Dow Agrosciences, 2012; DAS, 2014; Dow Agrosciences, 2014; Dow Agrosciences, 2015). The median field size was estimated to be 20, 15 and 20 acres for shallow shank, deep shank and drip applications, respectively. The generic seasonal air concentrations for these field sizes were 170, 62 and 56 $\mu g/m^3$ for shallow shank, deep shank and drip applications, respectively. The use of a nominal flux of 100 $\mu g/m^2$sec allows for scaling of the generic air concentrations to the seasonal application rate (Barry, 2015b). The scaling was done by using the ratio of the seasonal over maximal application rate. The seasonal (median) application rate was calculated as described previously in the Occupational Exposure section, and was 153, 327 and 103 lb 1,3-D/acre for shallow shank, deep shank and drip application, respectively. Thus, for shallow shank, deep shank and drip applications, the respective estimated seasonal air concentrations (SAC) were 78.3, 61.1 and 22.9 $\mu g/m^3$ or 0.0173, 0.0135 and 0.0050 ppm (Table IV.10).

Annual and lifetime air concentration

The assumption for residential bystander annual exposure is that a field will be fumigated only once a year according to the current agricultural practice. Accordingly, the bystander exposure to 1,3-D due to a single field fumigation is expected to be less important over a longer term (e.g., annual) and eventually be indistinguishable from the 1,3-D exposure due to ambient air. This expectation is consistent with the higher exposure estimates of STAC (up to a factor of ~7) and SAC (up to a factor of ~4) of residential bystanders at the edge of the buffer zone than ambient air. For this reason, the annual and lifetime exposures are not estimated in this section.

b. Residential bystander exposures to tree and vine applications

Short-term exposures for residential bystanders to tree and vine applications were calculated using air concentration data produced by Dow AgroSciences. Cryer and van Wesenbeeck used the ISCST model to simulate 20 years of 1,3-D air concentrations occurring 2 and 3 days after tree and vine applications (Cryer and van Wesenbeeck, 2000a; Cryer and Van Wesenbeeck, 2000b). The original air monitoring data were derived from fields treated with 12 gal 1,3-D per acre instead of 35 gal Telone II per acre (344 lbs 1,3-D/acre). Hence, the simulated air concentrations were scaled up using an adjustment factor to account for the difference in application rates. The 95^{th} percentile 24-hr modeled air concentrations were calculated for receptors located between 100 and 1000 feet from treated fields of various sizes (5.74-74.4 acres) (Powell, 2000). The ISCST3 modeling suggested that peak air concentrations occurred in the two to three day period following application. Larger treated fields were associated with higher air concentrations of 1,3-D (data not shown). The highest 24-hr air concentration modeled at the 100 ft buffer zone was 416.8 $\mu g/m^3$ (Powell, 2000) or 0.0918 ppm (Table IV.10).

Seasonal and annual residential bystander exposures to tree and vine applications were not determined due to the lack of long-term modeling of air concentrations. Lifetime exposures are not expected because of the limited potential for such exposures (orchards and vineyards are fumigated and replaced once in 20-30 years).

c. Residential bystander exposure from ambient air (measured air concentrations)

Several ambient air monitoring studies were performed in California between 1991 and 2006 (an extensive reference list can be found in (Wofford *et al.*, 2009). These studies monitored the 1,3-D ambient air concentrations during peak seasonal use. Accordingly, the monitoring results do not represent the yearly ambient air levels of 1,3-D needed for calculating long-term human exposures.

Two year-round studies of 1,3-D ambient air levels were conducted by DPR (in collaboration with the California Air Resources Board, CARB) and by the registrant, Dow AgroSciences (DAS). The first study was conducted in 2006 in Parlier, a small rural community in Fresno County (a high-use county for 1,3-D) (Wofford *et al.*, 2009). DPR conducted ambient air analysis for a number of pesticides, VOCs and other air pollutants in collaboration with CARB. CARB collected 24-hour 1,3-D samples every 6 days from January 17, 2006, through January 6, 2007. The sampling frequency increased to every three days during peak high-use periods of 1,3-dichloropropene. 1,3-D samples were collected at Benavidez School in Parlier. Within 6-7 miles of Parlier, the reported 1,3-D use was 302,075 lbs applied in 2006, suggesting a considerable exceedance of the township cap of 90,250 lbs. The acres treated, and number of reported applications were 934, and 122, respectively, in the same area. 71 samples of 1,3-D were collected, and 34% of these samples had quantifiable concentrations of 1,3-D. The limit of quantitation (LOQ) achieved in this study was 0.454 $\mu g/m^3$. The highest measured one-day concentration for 1,3-D was 23.6 $\mu g/m^3$ or 0.005 ppm. The one-year average concentration was 1.97 $\mu g/m^3$ or 0.0004 ppm.

The second study was conducted in Merced County in 2010-2012 by Dow AgroSciences (DAS) (Rotondaro and Van Wesenbeeck, 2012a). This study was detailed in the Occupational Bystander Exposure Estimates section. It employed 9 receptors in a variety of locations in 9 contiguous townships. Approximately 1300 continuous air samples were collected in the 14.5 months of the study, of which 72% were quantifiable (LOQ = 0.05 $\mu g/m^3$). The 1,3-D use in the 14.5 months of sampling ranged between 0 and 257,000 lbs 1,3-D in the nine monitored townships. With its robustness and sensitivity, this study was deemed appropriate for estimation of human bystander exposure to 1,3-D in ambient air, with the understanding that the township cap was exceeded up to 2.5 times in four of the 9 monitored townships.

The Merced Study indicated that the months of November and December exhibited high 1,3-D use, and the study duration of 14.5 months included two such periods. However, only the air concentrations measured from twelve consecutive months (January 2011-December 2011) were used for estimating residential bystander exposures.

Short-Term air concentration

The short-term residential bystander exposures from ambient air were based on the highest three-day air concentration of 1,3-D as recorded on December 14th, 2011 at Township #5 (township 07S11E). It was 369.2 $\mu g/m^3$ or 0.0813 ppm (Table IV.10).

Seasonal air concentration

The SAC exposure estimate for the residential bystander was calculated using the seasonal mean air concentration at Township #5. The length of the season was calculated by summing the number of consecutive months having application amounts equal to or greater than 5% of the annual total. The length of the season in Merced County was 4 months (September-December) (Fig. II.2.). The mean of the air concentrations measured during this use season is 20.42 $\mu g/m^3$ or 0.0045 ppm (Table IV.10).

Annual air concentration

As pointed out previously, the annual township cap was exceeded in several of the townships included in the Merced monitoring study. Consequently, an annual average derived from the Merced monitoring study is inappropriate for use in evaluating the health risk associated with annual exposure to 1,3-D. In this risk assessment, SOil Fumigant Exposure Assessment System (SOFEA), a computer model which allows a yearly application frequency that is consistent with the township cap limit, was used for generating the ambient annual air concentration. The SOFEA-2 modeling was performed by the registrant (see Section IV.B.3.d. Residential bystander exposure from ambient air [modeling]). The ambient air concentration used for characterizing the annual exposure of residential bystanders was 0.99 $\mu g/m^3$ or 0.0002 ppm; this value (i.e., median) was derived from 129,600 simulated annual ambient air concentrations of Township #5 (the highest measured 1,3-D concentrations in Merced County).

Lifetime air concentration

The LAC values for residential bystander exposure to ambient levels of 1,3-D were not calculated in this section. Multiple LAC values were generated in the following section for this exposure scenario using a computer modeling approach.

d. Residential bystander exposure from ambient air (modeling)

Lifetime Exposure

For estimating the lifetime exposure of residential bystanders, long-term ambient air concentrations of 1,3-D are not available. Hence, stimulated air concentrations coupled with stochastic (i.e., probabilistic) human exposure assessment models were used. The simulated air concentrations were generated from an air dispersion model for soil fumigant, SOil Fumigant Exposure Assessment System (SOFEA©) (Cryer *et al.*, 2004), based on the use patterns of 1,3-D in Merced, CA. SOFEA is a bystander exposure model developed by Dow AgroSciences (DAS), the registrant of soil fumigant 1,3-D (Cryer *et al.*, 2004). Technical description of the model description has been detailed elsewhere (van Wesenbeeck and Cryer, 2004; van Wesenbeeck *et al.*, 2011). Briefly, using the Industrial Source Complex Short-Term Model version 3 (ISCST3) as its computational engine, SOFEA simulates the 1,3-D concentrations in ambient air based on measured or simulated volatility losses from the treated field (i.e., flux profile). Also, Monte Carlo sampling, a stochastic technique, is employed to evaluate the effects of various parameters on the simulated 1,3-D air concentrations; these parameters include

weather, field size, application date, application rate, application type, depth, pesticide degradation rates in air, tarp presence, field re-treatment, and buffer setbacks.
The stochastic human exposure assessment models employed are High-End Exposure version 5, Crystal Ball (HEE5CB) (CDPR, 1997b) and Monte Carlo Annual-Based Lifetime Exposure model (MCABLE) (Driver *et al.*, 2015); both models were implemented with the latest version of Crystal Ball (release number 11.1.2.400 [32 bit]), an Excel Add-in program developed by Oracle Corporation, CA.

The HEE5CB model was developed by CDPR for use in the 1,3-D cancer risk assessment (CDPR, 1994; CDPR, 1997b). The technical details of the model have been described elsewhere (Sanborn and Powell, 1994; Johnson and Powell, 2005). Briefly, this model estimates the population lifetime exposure to 1,3-D (i.e., Lifetime Average Daily Dose [LADD]) from birth to age 30 or 70 based on a weighted average air concentration. The weighted average is calculated using air concentrations derived from five randomly selected locations within a high 1,3-D use area and the proportion of time an individual spent in each location. The proportion of time an individual spent in each location was derived from two surveys concerning daily activity patterns of California residents (Wiley, 1991; Wiley *et al.*, 1991) and was treated in the model as a stochastic multinomial variable. The goal of this model is to evaluate the exposure to 1,3-D by individuals based on the relative amount of time spent in a highest-exposure township and its surrounding townships within a high 1,3-D use areas.

The Monte Carlo Annual-Based Lifetime Exposure model (MCABLE) is a population based stochastic human exposure assessment model developed by DAS (Driver *et al.*, 2015). Similar to HEE5CB, LADD is calculated based on the air concentrations from five randomly selected locations within a high 1,3-D use area and the proportion of time an individual spent in each location. However, MCABLE also considers inputs including individual movement in and out of a high 1,3-D use area, time spent temporarily away from the high 1,3-D use area (i.e., "time-away"), and the change in residence within the area. Each of these inputs was sampled stochastically by the model from specific probability distributions developed by Driver *et al.* (2015) using a survey conducted in two high 1,3-D use areas in California: Merced and Ventura Counties (Kaplan, 2014).

Both HEE5CB and MCABLE models have inputs such as 1,3-D air concentration, breathing rate, body weight, daily time spent in each location within a high 1,3-D use area, and residency per lifetime in the area. Among these model inputs, sensitivity analysis indicates that 1,3-D air concentration and mobility-residency are associated with >90% of the uncertainties in the LADD calculations and therefore, judged to be the key factors affecting the cancer risk assessment of 1,3-D (DAS, 2015b; Driver *et al.*, 2015). The air concentration and residency-mobility are further discussed in the sections below. This will be followed by the description of exposure simulation results using the MCABLE and HEE5CB.

1,3-D Air Concentrations

As mentioned previously, for assessing the lifetime exposure to 1,3-D by residential bystanders, SOEFA was used to simulate the needed air concentrations. Since its inception in 2004, SOFEA Version 1 (i.e., SOFEA-1) has undergone modifications in programming and assumptions (USEPA, 2004; van Wesenbeeck *et al.*, 2014; Cryer *et al.*, 2015). The latest version of the program is called SOFEA Version 2 (SOFEA-2). Before it was submitted to CDPR, this version of SOFEA was further modified by incorporating updates including the air temperature input profile and mixing height (MH) (Cryer *et al.*, 2015). These updates were implemented in order to address comments by CDPR, which included the under predictions of measured 1,3-D concentrations at Merced, CA (Johnson, 2014a; Johnson, 2014b).

Based on the registrant's analysis, the inability of previous SOFEA models to capture the measured concentrations may have resulted from constant mixing height (MH) assumption and the extensive calms (<1 ms^{-1}) and variable low wind speeds prevalent in the winter (December/January) in Merced County (van Wesenbeeck *et al.*, 2015). Figure IV.3 shows an empirical quantile-quantile plot of simulated *versus* observed mean values of the 1,3-D concentrations (from January to December of 2011). These simulated air concentrations were generated by SOFEA-2 with a sinusoidal air temperature input profile and variable MH correction. As shown in Figure IV.3, the modified SOFEA-2 under-predicted 1,3-D air concentrations in 6 townships but over-predicted the air concentration in 3 townships. Figure IV.4 shows 9 empirical quantile-quantile plots of simulated versus observed 72-hr values of the 1,3-D concentrations. The SOFEA's predictions were lower than those observed in Township # 2, 5, and 6. These under-predictions of SOFEA appeared to occur mainly in the early and late fall (Figures IV.5 and IV.6), with the largest discrepancy (a factor of ~5) being observed in the month of December, i.e., winter (Figure IV.6). It should be noted, however, that when comparing one-year monitoring results to the simulation values, SOFEA-2 was run in the "validation mode." Under the validation mode, only finite model outputs (i.e., simulated values) were used for comparing the monitoring results observed at different spatial locations. Hence, it is plausible that the finite model outputs employed and the matching of spatial occurrences of the modeled and observed concentrations may have contributed to the under-predictions. That is, the high 1,3-D air concentrations predicted by the model may have occurred at different spatial locations than the monitoring stations. Building on this assertion, a simulation exercise was performed independently by the registrants (van Wesenbeeck *et al.*, 2015) and the Environmental Monitoring Branch of CDPR (Barry, 2015a). By relaxing the requirement of spatial matching and increasing the model outputs, SOFEA-2 was able to capture the highest observed air concentrations of 1,3-D.

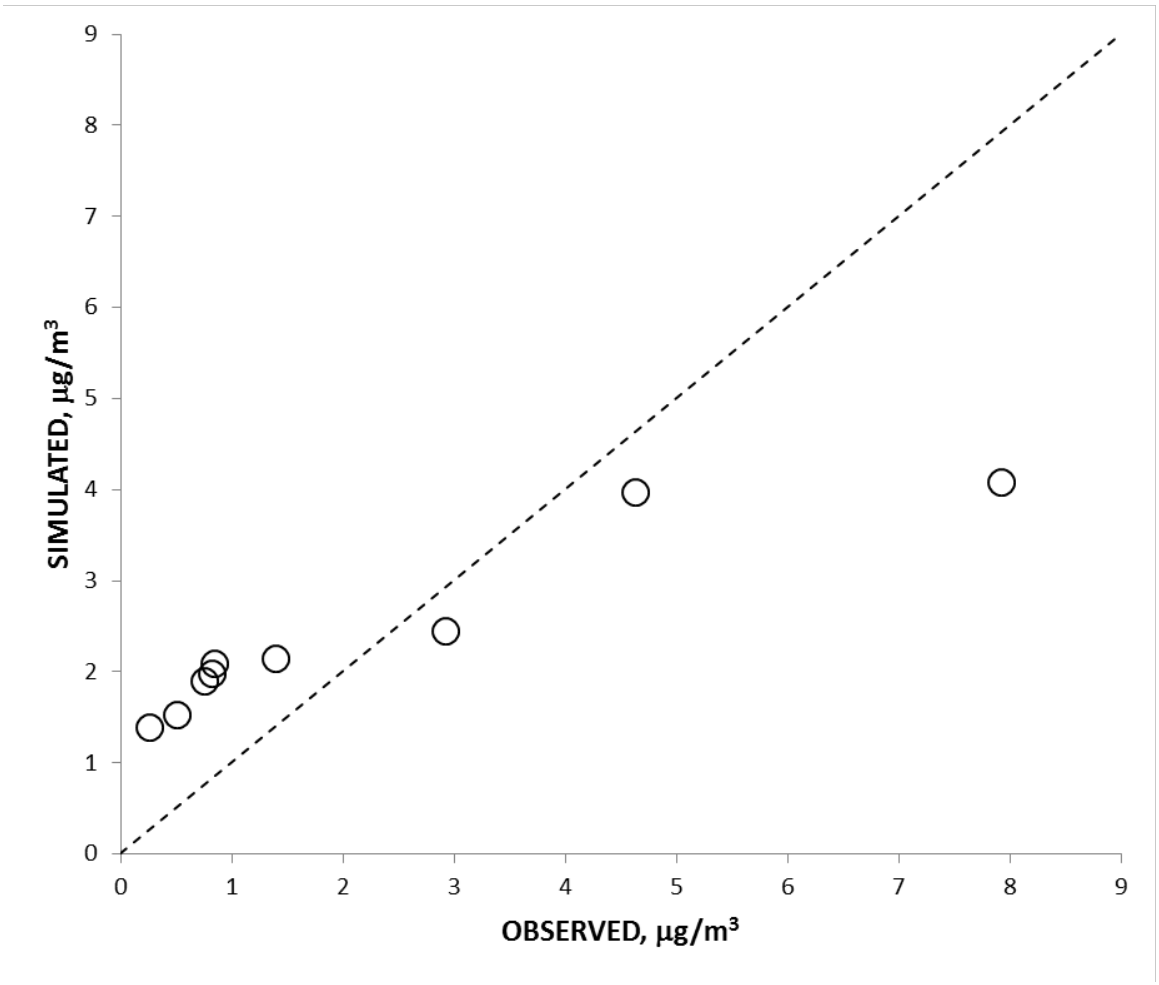


Figure IV. 2 Empirical quantile-quantile plot of the average annual concentrations (simulated *versus* observed) of 1,3-dichloropropene (1,3-D) in air at 9 contiguous townships of Merced County, CA, as performed in this risk assessment. The annual averages were calculated using the data from van Wesenbeeck and Cryer (2014). The dashed-line is the line of perfect prediction.

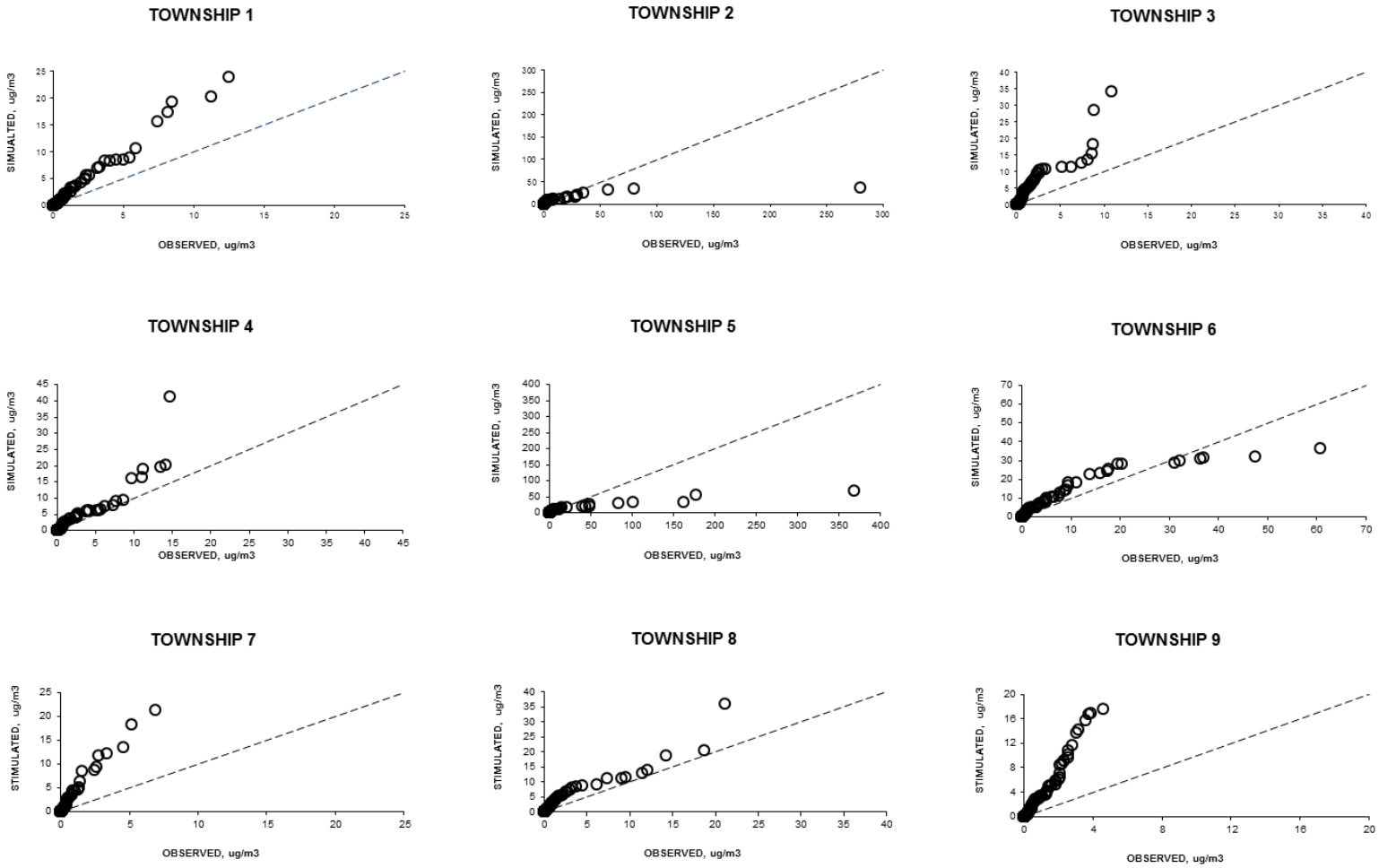


Figure IV. 3 Empirical quantile-quantile plots of the simulated versus observed 72-hour values of 1,3-dichloropropene (1,3-D) concentrations in air at 9 contiguous townships of Merced County, CA, as performed by this risk assessment. The data were obtained from van Wesenbeeck and Cryer (2014). The dashed-line is the line of perfect prediction.

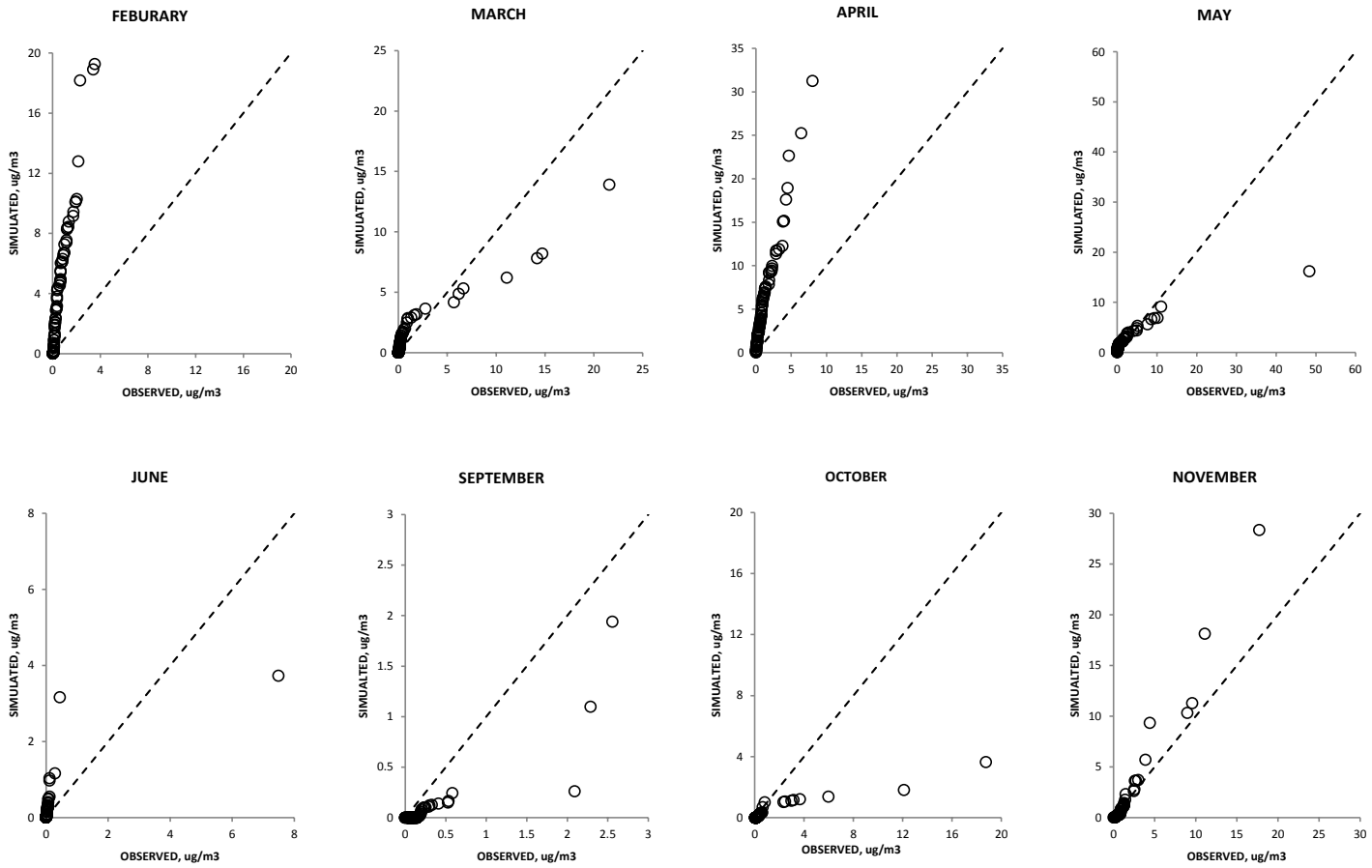


Figure IV. 4 Empirical quantile-quantile plots of the simulated versus observed 1,3-dichloropropene (1,3-D) concentrations in air (all-township combined). Only months with reported 1,3-D use are presented, as performed by this risk assessment. The data were obtained from van Wesenbeeck and Cryer (2014). The dashed-line is the line of perfect prediction.

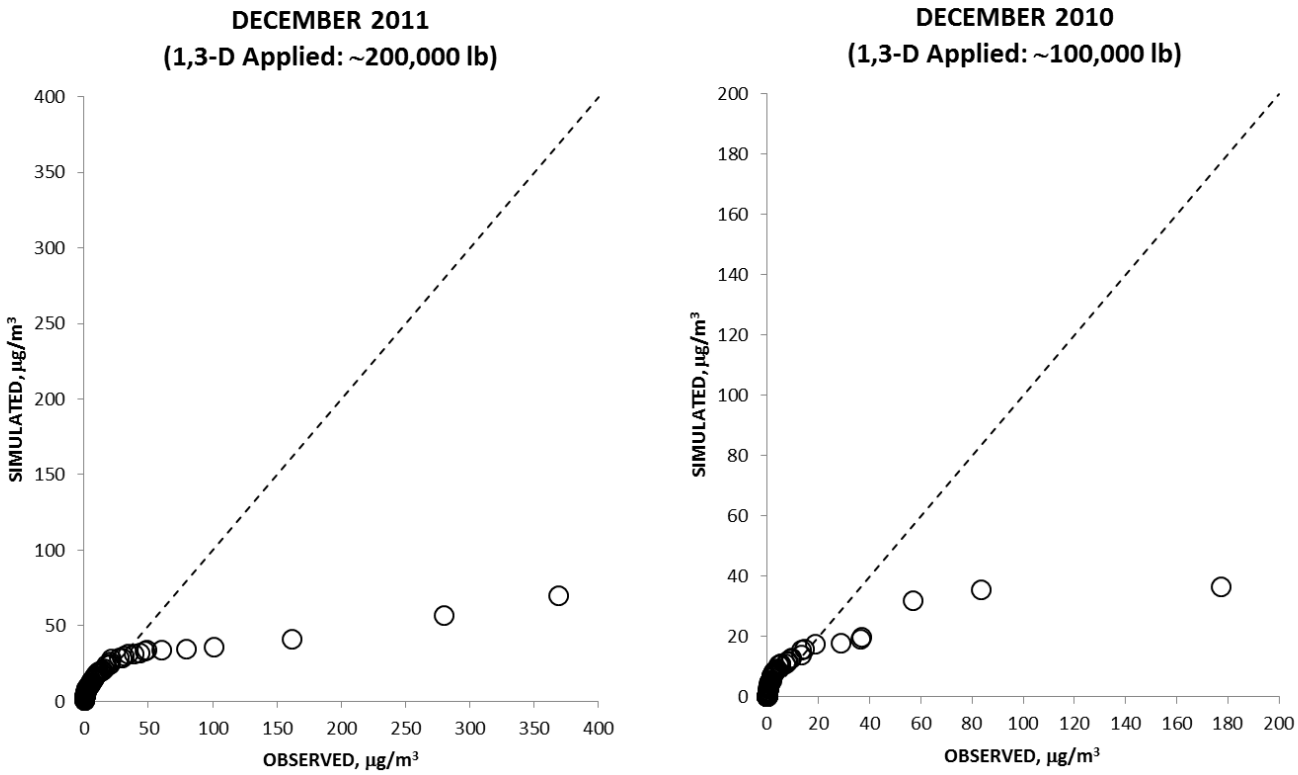


Figure IV. 5 Empirical quantile-quantile plots of simulated *versus* observed 1,3-dichloropropene (1,3-D) concentrations in air (all-township combined) in December of 2010 and 2011, as performed by this risk assessment. The 2010 values are presented for comparison. The dashed-line is the line of perfect prediction.

Residency-Mobility

As described previously, residency-mobility is one of the key factors affecting the lifetime exposure estimates of 1,3-D for residential bystanders. In this risk assessment, residency-mobility assessment includes the following: (1) individual movement in and out of a high 1,3-D use area, (2) relative amount of time spent in a highest-exposure township and its surrounding areas, and (or) (3) time spent temporarily away from the high 1,3-D use area (i.e., "time-away').

Detailed discussions of the assumptions employed by HEE5CB and MCABLE are beyond the scope of this document but can be found in two separate reports by CDPR (Sanborn and Powell, 1994) and the registrant (Driver *et al.*, 2015). Briefly, in the HEE5CB, two mobility assumptions are employed for estimating the lifetime average daily dose (LADD) (Johnson, 2007): low mobility and intermediate mobility. For the low mobility assumption, exposures are simulated based solely on the distribution of 1,3-D air concentrations from the highest-exposure township: Township #5. This setting is equivalent to stating that individuals spend their entire lives in Township #5. For the intermediate mobility assumption, the model employs air concentration distributions of 1,3-D from both the highest-exposure township (Township #5) and its surroundings (i.e., Townships #1, 2, 3, 4, 6, 7, 8, and 9). Under this intermediate mobility assumption, individuals are allowed to spend time (i.e., "move around") within five of the nine different townships; however, Township #5 is considered as "home" (i.e., an individual spends most of its time) and others four are considered as "away from home." Also, HEE5CB allows the time spent in these five townships to change with an individual's age. Although the actual time allocation among these townships are treated in HEE5CB as a stochastic multinomial variable, in general, it decreases from 80% for infants to about 60% for adults (Sanborn and Powell, 1994).

$$\text{LADD}=\left(\sum_{i=1}^{10} RT_i \left[\frac{Conc_i \text{ x } BR_i}{BW_i}\right]\right) \text{x } \frac{1}{70}$$

where the summation is over 10 age intervals,

RT_i = number of years in age interval *i* that the person resides in the high 1,3-D use area,

$Conc_i$ = annual average of air concentrations ($\mu g/m^3$) in 5 locations weighted by the proportion of time spent in each location in interval *i* ,

BR_i = average breathing rate (m^3/day) at each of 4 activity levels weighted by proportion of time spent at each level in interval *i*,

BW_i = body weight (kg) in interval *i*, and 70 years is the assumed lifetime.

When estimating lifetime exposure for use in the 1,3-D cancer risk assessment, like the HEE5CB, the MCABLE considers the relative amount of time that an individual spends in a highest-exposure township and its surrounding townships within a high 1,3-D use area.

However, the MCABLE also incorporates assumptions regarding (1) the age of an individual moved in and out of the area; (2) the fraction of individual's time temporary away from the high 1,3-D use area; (3) number of years that an individual resides in a single residence within the highest-exposure township before moving into other townships (up to three times) within the same area (Driver *et al.*, 2015). These additional variables in MCABLE are selected randomly from three separate custom distributions developed by Driver *et al.* (2015) based on a California-specific residential mobility survey (Kaplan, 2014). In that residential mobility survey, results indicate that, on average, individuals live ~25 years in a high 1,3-D use area (e.g., Merced, CA), with the 95^{th} and 99.9^{th} percentile residency values of ~50 years and >70 years, respectively (Driver *et al.*, 2014b). It should be noted that the residence of 25 years is similar to the recommended exposure duration (i.e., 30 years) for use in determining the individual cancer risk for a maximally exposed individual resident (OEHHA, 2012), and the residence of 70 years is the typical assumption of lifetime exposure duration for calculating cancer risk (USEPA, 2002). Also, it is noteworthy that adjustments for exposure to a carcinogen using the fraction of time spent among different townships within the high 1,3-D use area and time away from the high 1,3-D use area are consistent with the fact that exposure may be reduced when an individual moves within and outside the area (OEHHA, 2012).

In the MCABLE model, the average daily dose (ADD) is calculated for every year of a simulated individual's lifetime using the age- and gender-specific breathing rates and body weights together with an annual average of air concentration The annual average concentration is calculated using the air concentrations selected from 5 of the 9 townships weighted by the proportion of time spent by an individual in each township within the high 1,3-D use area. Also, the selection of a set of 5 air concentrations depends on whether an individual moves into another township (i.e., change in residence) within the same high 1,3-D use area. That is, if an individual moves from one township to another township, the air concentration of "old" township will be replaced by the "new" township for calculating the annual average air concentration. To accommodate the fact that individual may spend time temporary away the high 1,3-D use area (Kaplan, 2014), the ADD is adjusted by a "time-away" factor, F_a.

For calculating the LADD, the MCABLE considers both the exposures within and outside a high 1,3-D use area. The outside exposures (i.e., background) includes those occurred before and after an individual's residence in the high 1,3-D use area. By design, the MCABLE adds 100 years of ADD values; the sum is then divided by an individual's lifetime. The total number of years when the exposure occurred within the high 1,3-D use area is the difference between the age that an individual moves in (i.e., variable "i") and the age that an individual moves out (variable "n") of the area.

$$ADD = \left((1-F_a) \left[\frac{Conc_i \times BR_i}{BW_i} \right] \right)$$

$$LADD = \left[\left(\sum_i^n ADD_i \right) + Background \right] \times \frac{1}{lifetime}$$

where the summation is over a stochastically determined interval from i to n. For example, $\sum_{10}^{30} ADD$ would be equivalent to the assumption that an individual entered the community at the age of 10 (i.e., i = 10) and left at the age of 30 (i.e., n =30) after staying for 20 years.

F_a	=	fraction of time spent away from the high 1,3-D use area,
$Conc_i$	=	annual average of air concentrations ($\mu g/m^3$) in 5 of the 9 townships weighted by the proportion of time spent in each location in interval *i* ,
BR_i	=	age and gender specific average breathing rate (m^3/day) ,
BW_i	=	age and gender specific body weight (kg) in interval *i*,
Lifetime	=	70 years is the assumed lifetime for a male and a female[15],
Background	=	a value of total ADD due to the background exposure, calculated by adding ADD values from 100 – (n-i) simulation years.

Exposure Simulation Results

For simulating the lifetime exposures, SOFEA-2 modeling was performed by the registrant to obtain 1,3-D concentrations in air (Driver *et al.*, 2015; van Wesenbeeck *et al.*, 2015). Briefly, air concentrations of 1,3-D were generated based on the use scenarios (i.e., use rates and crop specific uses) and weather conditions in Merced, CA. After a round (i.e., "one year") of simulation, an average concentration of 1,3-D was generated at each of the 11,664 locations or receptors (i.e., 36 square mile/township x 36 receptors/square mile x 9 townships = 11,664 receptors over the entire area); the simulation was repeated 100 rounds (i.e., "100 years").

Using the stochastic exposure assessment models (i.e., MCABLE or HEE5CB), an annual lifetime average daily exposure was estimated based on the list of 100 x 11,644 annual average concentrations. Distribution of the exposure estimates was generated by Latin hypercube sampling method with 10,000 trials. With few exceptions (see below), modeling settings described by Driver *et al.* (2015) for MCABLE and Sanborn and Powell (1994) for HEE5CB were followed.

[15]The original lifespan assumption of 75 years for a male and 80 years for a female in the MCABLE are replaced by 70 years (both sexes); 70-year is the typical lifetime exposure duration for use in calculating cancer risk (USEPA, 2002).

Table IV.11 shows the estimates of lifetime average daily dose (LADD) of individuals living (1) variable time of, (2) 30 years of, (3) 50 years of, or (4) 70 years of a total 70-year exposure/lifetime in a high 1,3-D use area using MCABLE. In the MCABLE model, the duration of time spent and the age of arrival ("start age") in the high use area are treated as stochastic variables. These settings are equivalent to stating that the total number of years staying and the time of moving in (and out) by an individual are not "fixed" and vary within a population (i.e., not duration-specific). For simulating a duration-specific exposure, the MCABLE model was modified such that the total length of time spent in the high 1.3-D use area was restricted to 30, 50, or 70 years, and only the "start age" was varied stochastically (see RESIDENCY-MOBILITY section for the rationale of selecting these time intervals). This is equivalent of assuming that an individual moves into the high use area at any time of their lifetime but stays there for a fixed period. Using the build-in function of MCABLE, the effect of "time-away" from the area on 1,3-D exposure estimate was also evaluated.

To further evaluate the effect of residence on exposure to 1,3-D, LADD was also estimated separately using HEE5CB. In this risk assessment, the original mobility assumptions of HEE5CB were expanded to three: low, intermediate, and high. Also, under each mobility assumption, individuals were assumed to spend the time from birth to age 30, 50, or 70. At a given time spent, exposure was estimated using the air concentration distributions from (1) the highest exposure township (i.e., low mobility), (2) the highest exposure township plus its surrounding townships (intermediate mobility), or (3) all townships (high mobility). Township #5 was selected as the highest exposure township based on the air monitoring results of Rotondaro and van Wesenbeeck (2012b). Comparing to MCABLE, the mobility assumptions in HEE5CB are more restrictive (Driver *et al.*, 2015). However, results of the mobility survey by Kaplan (2014) indicated the existence of individuals who spent ≥30-year (36.8%) , ≥50-year (13.8%), or ≥ 70-year (2.1%) durations in Merced, CA, where high use of 1,3-D occurred (Kaplan, 2014). Hence, the exposure scenarios of HEE5CB can be considered as the special (or limiting) cases of MCABLE's. It is noteworthy that because the mobility assumption of HEE5CB dictates that the total number of years staying and the time of moving in starting from birth, the higher age-specific breathing rates and lower body weights of children than adults are expected to be major contributing factors to the higher LADD values generated by HEE5CB than MCABLE (Table IV.11).

Table IV. 11 Lifetime Average Daily Dose (μg/kg/day) of 1,3-D Inhaled by Residential Bystanders in a High 1,3-D Use Area.

Residence Time	MCABLE[a]			
	Male		**Female**	
	Mean	**95th Percentile**	**Mean**	**95th Percentile**
With Time-Away				
Variable	0.0960	0.1887	0.0880	0.1778
30-Year Fixed	0.1007	0.1752	0.0927	0.1644
50-Year Fixed	0.1372	0.2338	0.1254	0.2168
70-Year Fixed	0.1791	0.3056	0.1631	0.2833
Without Time-Away				
Variable	0.1064	0.2052	0.0976	0.1918
30-Year Fixed	0.1109	0.1880	0.1025	0.1779
50-Year Fixed	0.1532	0.2574	0.1387	0.2366
70-Year Fixed	0.1999	0.3326	0.1819	0.3048
Residence Time	**HEE5CB[b]**			
	Male		Female	
	Mean	95th Percentile	Mean	95th Percentile
High Mobility				
Birth to age 30	0.2028	0.3467	0.1976	0.3393
Birth to age 50	0.2925	0.4999	0.2799	0.4828
Birth to age 70	0.3823	0.6554	0.3623	0.6265
Intermediate Mobility				
Birth to age 30	0.2301	0.4200	0.2724	0.4982
Birth to age 50	0.3609	0.6433	0.3189	0.5830
Birth to age 70	0.4246	0.7695	0.4122	0.7569
Low Mobility				
Birth to age 30	0.2568	0.4466	0.3087	0.5518
Birth to age 50	0.4218	0.7453	0.3557	0.6307
Birth to age 70	0.4850	0.8396	0.4605	0.8183

[a] Average daily dose (ADD) was estimated using a set of five air concentrations selected randomly from a list of 100 x 11,644 average annual 1,3-D concentrations produced by SOFEA-2 (Driver *et al.*, 2015). The LADD was determined by averaging the total ADD using an individual's lifetime of 70 years.

[b] LADD was estimated using a set of five cumulative probability distributions of average annual concentrations as described in Sanborn and Powell (1994). These cumulative probability distributions were derived from a list of 31 x 11,644 average annual 1,3-D concentrations produced by SOFEA-2. The selection criterion of these 31 lists is further detailed in EXPOSURE APPRAISAL section. For a given mobility assumption, multiple simulations were conducted but only the highest LADD was presented.

C. Risk characterization

The potential for non-oncogenic health effects resulting from exposure to 1,3-D was expressed as the margin of exposure (MOE). An MOE is the ratio of the BMCL or NOEL value derived from the definitive acute, subchronic or chronic studies, divided by the estimated human exposure value. As this assessment was focused on risks emanating from inhalation exposure to 1,3-D vapor, both the BMCL (or NOEL) and the exposure values are expressed as air concentrations (ppm) rather than as internal doses (mg/kg).

Margin of Exposure (MOE) = BMCL or NOEL (in ppm) / Exposure dose (in ppm)

For adults under occupational or non-occupational exposure conditions, MOEs of 30 were considered adequate to protect human health. This "target MOE" was the product of an uncertainty factor of 3, to account for pharmacodynamic differences between laboratory animals and humans [16], and 10, to account for an assumed 10-fold range of sensitivity within the human population. For children, who are presumably exposed only under non-occupational scenarios, MOEs of 100 were considered to be health protective. The extra ~3-fold factor was due to database uncertainty arising because no toxicity studies were conducted on young animals. Consequently, we had no way of assessing the possibility that infants and children might be more susceptible to the toxic effects of 1,3-D.

1. Occupational exposure scenarios

a. Acute / short-term risk: occupational scenarios

MOEs for acute / short-term occupational scenarios were calculated by applying the critical occupational HEC of 33 ppm to the exposure scenarios indicated in section IV.B. The resultant values are listed below in Table IV.12. The highest acute / short-term risk occupation was tarp remover, which showed MOEs of 1 for shallow shank, deep shank and drip applications. Two additional occupational scenarios registered MOEs below the target MOE of 30, including applicator (injection auger) and occupational bystander (shallow shank without tarp). Occupational bystander (drip with tarp) showed an MOE of exactly 30.

[16] The additional 3x uncertainty factor for pharmacokinetic differences between animals and humans was considered accounted for by the use of the RGDR approach to estimate human equivalent concentrations.

Table IV. 12 1,3-D exposure estimates and resultant MOE values, occupational scenarios: acute / short-term toxicity

Exposure scenario	Air concentration (ppm)	HEC (ppm) [a]	MOE	Target MOE [b]
Acute / short-term exposures				
Applicator (shallow shank w/o tarp)	0.27	33	122	30
Applicator (shallow shank with tarp)	0.85	33	39	30
Applicator (deep shank w/o tarp	0.27	33	122	30
Applicator (deep shank with tarp)	0.85	33	39	30
Applicator (drip w/o tarp)	0.28	33	118	30
Applicator (drip with tarp)	0.23	33	143	30
Applicator (injection auger)	1.2	33	28	30
Loader (shallow shank)	0.70	33	47	30
Loader (deep shank)	0.70	33	47	30
Tarp remover (shallow shank)	33	33	1	30
Tarp remover (deep shank)	33	33	1	30
Tarp remover (drip)	33	33	1	30
Reentry worker (shallow shank)	0.037	33	892	30
Reentry worker (deep shank)	0.037	33	892	30
Reentry worker (drip)	0.037	33	892	30
Occupational bystander (shallow shank w/o tarp)	2.0	33	17	30
Occupational bystander (deep shank w/o tarp)	0.6	33	55	30
Occupational bystander (drip with tarp)	1.1	33	30	30

[a] HECs were calculated above in section IV.A.1.b.

[b] Target MOEs were determined by multiplying the uncertainty factors for intra- and interhuman variability (see Table IV.2.a).

b. Subchronic / seasonal risk: occupational scenarios

MOEs for subchronic / seasonal occupational scenarios were calculated by applying the critical occupational HEC of 0.90 ppm to the exposure scenarios indicated in section IV.B. The resultant values are listed below in Table IV.13. The highest seasonal risk occupation was tarp remover (deep shank), which showed an MOE of 0.11. Several additional occupational scenarios registered MOEs below the target MOE of 30, including tarp remover (shallow shank; MOE = 0.23), tarp remover (drip; MOE = 0.35), applicator (shallow shank without tarp; MOE = 28), applicator (shallow shank with tarp; MOE = 9), applicator (deep shank without tarp; MOE = 13), applicator (deep shank with tarp; MOE = 4), applicator (drip without tarp; MOE = 23), loader (shallow shank; MOE = 15), loader (deep shank; MOE = 7), and reentry worker (deep shank; MOE = 28).

Table IV. 13 1,3-D exposure estimates and resultant MOE values, occupational scenarios: subchronic / seasonal risk

Exposure scenario	Air concentration (ppm)	HEC (ppm)	MOE	Target MOE
Subchronic / seasonal exposures				
Applicator (shallow shank w/o tarp)	0.032	0.90	28	30
Applicator (shallow shank with tarp)	0.10	0.90	9	30
Applicator (deep shank w/o tarp	0.068	0.90	13	
Applicator (deep shank with tarp)	0.22	0.90	4	30
Applicator (drip w/o tarp)	0.039	0.90	23	30
Applicator (drip with tarp)	0.018	0.90	50	30
Applicator (injection auger)	n/a	0.90	n/a	30
Loader (shallow shank)	0.062	0.90	15	30
Loader (deep shank)	0.13	0.90	7	30
Tarp remover (shallow shank)	3.9	0.90	0..23	30
Tarp remover (deep shank)	8.3	0.90	0.11	30
Tarp remover (drip)	2.6	0.90	0.35	30
Reentry worker (shallow shank)	0.015	0.90	60	30
Reentry worker (deep shank)	0.032	0.90	28	30
Reentry worker (drip)	0.010	0.90	90	30
Occupational bystander (shallow shank w/o tarp)	0.0012	0.90	750	30
Occupational bystander (deep shank w/o tarp)	0.0012	0.90	750	30
Occupational bystander (drip with tarp)	0.0012	0.90	750	30

c. Chronic / annual risk: occupational scenarios

MOEs for chronic / annual occupational scenarios were calculated by applying the critical occupational HEC of 0.59 ppm to the exposure scenarios indicated in section IV.B. The resultant values are listed below in Table IV.14. The highest annual risk occupation was tarp remover (deep shank), which showed an MOE of 0.11. Several additional occupational scenarios registered MOEs below the target MOE of 30, including tarp remover (shallow shank; MOE = 0.49), tarp remover (drip; MOE = 0.69), applicator (shallow shank with tarp; MOE = 18), applicator (deep shank w/o tarp; MOE = 14), applicator (deep shank with tarp; MOE = 4), loader (deep shank; MOE = 7) and reentry worker (deep shank; MOE = 25).

Table IV. 14 1,3-D exposure estimates and resultant MOE values, occupational scenarios: chronic / annual risk

Exposure scenario	Air concentration (ppm)	HEC (ppm)	MOE	Target MOE
Chronic / annual exposures				
Applicator (shallow shank w/o tarp)	0.0096	0.59	61	30
Applicator (shallow shank with tarp)	0.032	0.59	18	30
Applicator (deep shank w/o tarp	0.042	0.59	14	30
Applicator (deep shank with tarp)	0.14	0.59	4	30
Applicator (drip w/o tarp)	0.013	0.59	45	30
Applicator (drip with tarp)	0.0060	0.59	98	30
Applicator (injection auger)	n/a	0.59	n/a	30
Loader (shallow shank)	0.019	0.59	31	30
Loader (deep shank)	0.082	0.59	7	30
Tarp remover (shallow shank)	1.2	0.59	0.49	30
Tarp remover (deep shank)	5.2	0.59	0.11	30
Tarp remover (drip)	0.85	0.59	0.69	30
Reentry worker (shallow shank)	0.0064	0.59	92	30
Reentry worker (deep shank)	0.024	0.59	25	30
Reentry worker (drip)	0.0044	0.59	134	30
Occupational bystander (shallow shank w/o tarp)	0.00062	0.59	952	30
Occupational bystander (deep shank w/o tarp)	0.00062	0.59	952	30
Occupational bystander (drip with tarp)	0.00062	0.59	952	30

d. Oncogenic risk: occupational scenarios

Oncogenic risk was calculated assuming both a portal of entry and systemic mode of action (Table IV.15). For portal of entry, the risk values were calculated using the upper confidence limit (UCL) air unit risk value of 0.0059 ppm^{-1} to characterize the dose-response relation between 1,3-D concentration and bronchioloalveolar tumor incidence for occupational scenarios. For systemic, the risk values were calculated using the UCL air unit risk value of 0.020 ppm^{-1}. All of the work tasks examined showed oncogenic risk values that exceeded the "negligible oncogenic risk" standard of $1x10^{-6}$, regardless of the assumed mode of action. Actual risk values for portal of entry ranged between 1.9 $x10^{-6}$ (occupational bystander, shallow and deep shank without tarp, and occupational bystander, drip with tarp) and $1.7x10^{-2}$ (tarp remover, deep shank). Risk values for systemic ranged between $6.6x10^{-6}$ (occupational bystander, shallow and deep shank without tarp, and occupational bystander, drip with tarp) and $5.6x10^{-2}$ (tarp remover, deep shank).

Table IV. 15 1,3-D exposure estimates and resultant oncogenic risk values: occupational scenarios assuming both portal of entry and systemic modes of action

		AUR (ppm^{-1}) [a]		Oncogenic risk		
Exposure scenario	**Air conc. (ppm)**	**Portal of entry**	**Systemic**	**Portal of entry**	**Systemic**	**Target onco. risk [b]**
Applicator (shallow shank w/o tarp)	0.0054	0.0059	0.020	$3.2x10^{-5}$	$1.1x10^{-4}$	$1x10^{-6}$
Applicator (shallow shank with tarp)	0.017	0.0059	0.020	$1.0x10^{-4}$	$3.4x10^{-4}$	$1x10^{-6}$
Applicator (deep shank w/o tarp	0.023	0.0059	0.020	$1.4x10^{-4}$	$4.6x10^{-4}$	$1x10^{-6}$
Applicator (deep shank with tarp)	0.072	0.0059	0.020	$4.3x10^{-4}$	$1.4x10^{-3}$	$1x10^{-6}$
Applicator (drip w/o tarp)	0.0070	0.0059	0.020	$4.1x10^{-5}$	$1.4x10^{-4}$	$1x10^{-6}$
Applicator (drip with tarp)	0.0032	0.0059	0.020	$1.9x10^{-5}$	$6.4x10^{-5}$	$1x10^{-6}$
Applicator (injection auger)	n/a	0.0059	0.020	n/a	n/a	$1x10^{-6}$
Loader (shallow shank)	0.0100	0.0059	0.020	$5.9x10^{-4}$	$2.0x10^{-4}$	$1x10^{-6}$
Loader (deep shank)	0.044	0.0059	0.020	$2.6x10^{-4}$	$8.8x10^{-4}$	$1x10^{-6}$
Tarp remover (shallow shank)	0.66	0.0059	0.020	$3.9x10^{-3}$	$1.3x10^{-2}$	$1x10^{-6}$
Tarp remover (deep shank)	2.8	0.0059	0.020	$1.7x10^{-2}$	$5.6x10^{-2}$	$1x10^{-6}$
Tarp remover (drip)	0.46	0.0059	0.020	$2.7x10^{-3}$	$9.2x10^{-3}$	$1x10^{-6}$
Reentry worker (shallow shank)	0.0034	0.0059	0.020	$2.0x10^{-5}$	$6.8x10^{-5}$	$1x10^{-6}$
Reentry worker (deep shank)	0.013	0.0059	0.020	$7.67x10^{-5}$	$2.6x10^{-4}$	$1x10^{-6}$
Reentry worker (drip)	0.0024	0.0059	0.020	$1.4x10^{-5}$	$4.8x10^{-5}$	$1x10^{-6}$
Occupational bystander (shallow shank w/o tarp)	0.00033	0.0059	0.020	$1.9x10^{-6}$	$6.6x10^{-6}$	$1x10^{-6}$
Occupational bystander (deep shank w/o tarp)	0.00033	0.0059	0.020	$1.9x10^{-6}$	$6.6x10^{-6}$	$1x10^{-6}$
Occupational bystander (drip with tarp)	0.00033	0.0059	0.020	$1.9x10^{-6}$	$6.6x10^{-6}$	$1x10^{-6}$

[a] The air unit risk was determined as described in section IV.A.3.c.

[b] Target oncogenic risk values were set at the generally accepted "negligible oncogenic risk" value of $1x10^{-6}$.

2. Non-occupational (residential bystander) exposure scenarios

a. Acute / short-term risk: non-occupational scenarios

Acute / short-term non-occupational MOEs were calculated by applying the critical HEC value of 11 ppm to the exposure estimates provided in section IV.B. They appear below in Table IV.16. The lowest MOE of 20 was determined for a resident / bystander at the edge of a buffer zone for a shallow shank application to an 80-acre site. This value was below both the adult and child target MOEs of 30 and 100, respectively. No other MOEs were below 30. However, two of the remaining scenarios---near application site, deep shank and drip applications---showed MOEs below 100. Ambient exposures generated an MOE of 135. These results appear in Table IV.16

Table IV. 16 1,3-D exposure estimates and resultant MOE values, non-occupational scenarios: acute / short-term toxicity

Exposure scenario	Air concentration (ppm)	HEC – adult (ppm) [a]	MOE	Target MOE – adult [b]	Target MOE – child [b]
Acute / short-term exposures					
Near application site, edge of buffer zone, shallow shank	0.5508	11	20	30	100
Near application site, edge of buffer zone, deep shank	0.1432	11	96	30	100
Near application site, edge of buffer zone, drip	0.1800	11	61	30	100
Near application site, edge of buffer zone, tree & vine	0.0918	11	120	30	100
Ambient	0.0813	11	135	30	100

[a] HECs were calculated above in section IV.A.1.b.

[b] Target MOEs were determined by multiplying the uncertainty factors for intra- and inter-human variability (see Table IV.4.

b. Subchronic / seasonal risk: non-occupational scenarios

MOEs for subchronic / seasonal risk were calculated by applying the critical HEC value of 0.30 ppm to the exposure estimates provided in section IV.B. The lowest MOE of 17 was determined for a resident / bystander at the edge of a buffer zone for a shallow shank application. This value was below both the adult and child target MOEs of 30 and 100, respectively. One other seasonal exposure scenario---near an applications site at the edge of a buffer zone, deep shank---showed a sub-30 MOE (MOE = 22). Exposures occurring near an application site at the edge of the buffer zone, drip (MOE=60) and ambient exposure (MOE=67) indicated a seasonal health risk to children. These results appear in Table IV.17.

Table IV. 17 1,3-D exposure estimates and resultant MOE values, non-occupational scenarios: subchronic / seasonal risk

Exposure scenario	Air concentration (ppm)	HEC – adult (ppm) [a]	MOE	Target MOE – adult [b]	Target MOE – child
Subchronic / seasonal exposures					
Near application site, edge of buffer zone, shallow shank	0.0173	0.30	17	30	100
Near application site, edge of buffer zone, deep shank	0.0135	0.30	22	30	100
Near application site, edge of buffer zone, drip	0.0050	0.30	60	30	100
Near application site, edge of buffer zone, tree & vine	nd				
Ambient	0.0045	0.30	67	30	100

[a] HECs were calculated above in section IV.A.1.b.

[b] Target MOEs were determined by multiplying the uncertainty factors for intra- and interhuman variability (see Table IV.4.

c. Chronic / annual risk (non-oncogenic): non-occupational scenarios

Chronic / annual exposure was not anticipated for non-occupational bystanders to specific 1,3-D applications. However, chronic / annual ambient exposure was expected. The MOE for chronic / annual ambient risk was calculated by applying the critical HEC value of 0.20 ppm to the ambient exposure estimate provided in section IV.B (Table IV.11). The MOE of 1000 did not indicate a health risk under ambient exposure conditions for adults or children. These results appear in Table IV.18.

Table IV. 18 1,3-D exposure estimates and resultant MOE values, chronic / annual risk, ambient exposure

Exposure scenario	Air concentration (ppm)	HEC – adult (ppm) [a]	MOE	Target MOE – adult [b]	Target MOE – child [b]
Chronic / annual exposures					
Ambient	0.0002	0.20	1000	30	100

[a] The chronic HECs was calculated above in section IV.A.1.b.

[b] Target MOEs were determined by multiplying the uncertainty factors for intra- and interhuman variability (see Table IV.4.

d. Oncogenic risk: ambient exposure scenarios

In this assessment, the oncogenic risks of various agricultural workers and occupational bystanders from 1,3-D exposure were characterized using the respective estimated lifetime air concentrations (LAC) in part-per-million (ppm) and an upper bound air unit risk value (ppm^{-1}). The air unit risk value was derived using species-specific minute volumes (*i.e.*, breathing rates) and surface areas of various respiratory tract regions in adult animals and humans. However, in order to evaluate the total lifetime oncogenic risk of a population, age-specific air unit risk values of 1,3-D were required since various constituent subpopulation, especially infants and children, were included. As detailed in section IV.C. (Risk Characterization), the requisite morphometric information---in particular, respiratory tract surface areas for children and immature mice---was not available. Consequently, these age specific air unit risk values were not calculated. As an alternative, an approach based on internal doses in mg/kg/day coupled with a 1,3-D cancer potency factor $(mg/kg/day)^{-1}$ was used (see Appendix VIII for the derivation of the cancer potency factor from the air unit risk values generated for this document). Each internal dose was calculated using default age-specific inhalation rates and body weights. Because 1,3-D is not a reproductive or developmental toxicant by the inhalation route (section IV.A., Hazard Identification), this alternative approach was used to integrate the effects of life-stage specific physiology in the evaluation of a population's total lifetime cancer risk.

Table IV.19 shows the estimated lifetime oncogenic risks at the 95^{th} percentile based on the cancer risk distributions generated by MCABLE or HEE5CB. As can be seen in Table IV.19, the oncogenic risk (regardless of the stochastic model employed) increases with increasing amount of time spent in a high 1,3-D use area. At a given residence time, time-away appears to have a minor impact on the oncogenic risk estimates by MCABLE. Because of the more restrictive mobility assumptions employed, oncogenic risks derived from HEE5CB are higher than those by MCABLE as expected.

Table IV. 19 Potential lifetime oncogenic risk (95th percentile) from inhaled 1,3-D in residential bystanders in a high 1,3-D use area [a]

Residence Time	MCABLE	
	Male	Female
	95th%-tile oncogenic risk x 10^6	
With Time-Away		
Variable	2.64-9.09	2.49-8.56
30-Year Fixed	2.45-8.44	2.30-7.91
50-Year Fixed	3.27-11.26	3.04-10.44
70-Year Fixed	4.28-14.72	3.97-13.64
Without Time-Away		
Variable	2.87-9.88	2.69-9.23
30-Year Fixed	2.63-9.06	2.49-8.57
50-Year Fixed	3.60-12.40	3.31-11.39
70-Year Fixed	4.66-16.02	4.27-14.68
Residence Time	**HEE5CB**	
	Male	Female
	95th%-tile oncogenic risk x 10^6	
High Mobility		
Birth to age 30	4.85-16.70	4.75-16.34
Birth to age 50	6.99-24.08	6.76-23.25
Birth to age 70	9.18-31.56	8.77-30.17
Intermediate Mobility		
Birth to age 30	5.88-20.22	6.97-23.99
Birth to age 50	9.01-30.98	8.16-28.08
Birth to age 70	10.77-37.06	10.6-36.45
Low Mobility		
Birth to age 30	6.25-21.51	7.73-26.57
Birth to age 50	10.43-35.89	8.83-30.38
Birth to age 70	11.75-40.44	11.46-39.41

[a] 1,3-D cancer risk was the 95th percentile value derived from the cancer risk distribution generated by MCABLE or HEE5CB. The individual risk value that constituted the cancer risk distribution was calculated as the LADD (μg/kg/day) times the human cancer potency factors: (a) 0.000014 $[\mu g/kg/day]^{-1}$ for portal-of-entry effect and (b) 0.000048 $[\mu g/kg/day]^{-1}$ for systemic effect developed in this risk assessment. For each range, the portal-of-entry estimate is the first (lower) value and the systemic estimate is the second (higher) value.

V. RISK APPRAISAL

Risk assessment is the process by which the toxicity of a chemical is compared to the potential for human exposure under specific conditions in order to estimate the risk to human health. Every risk assessment has inherent limitations relating to the relevance and quality of the toxicity and exposure data. Assumptions and extrapolations are incorporated into the hazard identification, dose-response assessment and exposure-assessment processes, resulting in uncertainty in the risk characterization, which integrates the information from those three processes. Qualitatively, risk assessments for all chemicals have similar uncertainties. However, the magnitude of those uncertainties varies with the availability and quality of the toxicity and exposure data, and with the relevance of that data to the anticipated exposure scenarios.

In the following sections, the uncertainties associated with characterization of health risks from exposure of workers and the general public to 1,3-D vapor are described. The exposure scenarios examined include only inhalation exposure to workers and to the general public.

A. HAZARD IDENTIFICATION

1. Acute / short-term risk

This analysis uses decrements in body weight gain in rats and mice sustained over several days of inhalation exposure to 1,3-D as the toxicologic drivers for the acute / short-term assessment. The study that yielded the most defensible animal BMCL and HEC values was the 13-week rat inhalation study of (Stott *et al.*, 1984). By study day 3, both males and females showed statistically significant decrements in body weight at 90 ppm compared to controls, reflecting decrements in weight gain during that period (Table III.3). Actual body weight losses occurred at 150 ppm.

Benchmark concentration (BMC) modeling of the day -1 to day 3 body weight data over the whole dose range yielded BMCLs of 49 and 51 ppm for males and females, respectively. These fell in the middle of the BMCL range for five studies which yielded statistically appropriate data (40-66 ppm). The BMCL value of 49 ppm was chosen to evaluate acute / short-term risk because the 3-day exposure was closest to an actual acute inhalation scenario. It is noted, however, that one study (Coate, 1979) produced a BMCL of 6 ppm, notably lower than those established in the other studies. While this study was not used to characterize acute / short term risk due to the distinct possibility that it was an outlier, it does serve to emphasize that choice of 49 ppm to characterize acute / short term risk may underestimate toxicity. In addition, John (1983) supplied evidence in a rat developmental toxicity study that very slight weight decrements were present even at 20 ppm. While this observation was an important caveat to the BMC data, it was not sufficiently robust to drive the acute assessment. We opted to use BMC modeling instead of the traditional NOEL / LOEL approach for the three reasons outlined by Poet (2010). First, the BMC approach is not constrained by the particular air concentrations used in the study,

especially the NOEL and LOEL levels. Instead, BMC modeling takes advantage of the entire dose range in order to establish the curve that most accurately defines the response at low concentrations. Second, BMC modeling avoids the imprecision inherent in establishing NOELs and LOELs since those values can theoretically fall at any infinitesimal point between the experimental values. And third, unlike the traditional approach, BMC modeling takes into account the number of animals tested per dose in order to define confidence limits. Studies with high animal numbers have tighter confidence limits, meaning that the lower bound (*i.e.*, the BMCL) is closer to the BMC. In essence, studies with high statistical power are rewarded with more accurate points of departure.

The actual BMC and BMCL values were obtained using a benchmark response of one standard deviation ($BMR_{1\delta}$) as recommended by USEPA for continuous data (USEPA, 2000). Three dose response models were tested---linear, polynomial and exponential---with best fit determined by Akaike's information constant, goodness of fit p value and BMC / BMCL ratio. Uncertainty associated with this type of modelling was inevitable since the accuracy of the BMCL values was a function of how well the curve fit the experimental data. Thus the critical BMCL could have over- or underestimated the theoretically "correct" value, with resultant uncertainty in the MOE calculation, though the narrow range of BMCLs determined for the 5 rodent studies made it unlikely that the critical BMCL of 49 ppm was far off the mark.

Use of bodyweight decrement as a critical driver in this risk assessment was accompanied by significant uncertainty, particularly with regard to the question of whether the observed weight decrements were of sufficient adversity to drive an acute / short-term health assessment. The operative assumption is that the animals emerged from the daily inhalation exposures with mild systemic illness rendering them uninterested or incapable of consuming as much food as unexposed controls. Lowered food consumption during the first week was evident in the rat studies of Gollapudi (1998) and John (1983).

An alternate possibility exists, however. In the concentration range identified by the BMC modeling, the animals could have smelled the 1,3-D, considered it noxious, and thus curtailed food consumption during the first few days of exposure. The human odor threshold for Telone II vapor was determined to be 4.4±3.1 ppm (Rick and McCarty, 1988), well below most of the rodent BMCL values. As the exposures continued, the animals adapted to the 1,3-D-containing air and began eating at control rates. Clearly, if the 1,3-D concentration was very high, the animals became physiologically compromised. In the 10-week rat dominant lethal study of (Gollapudi *et al.*, 1998), 60 ppm and 150 ppm animals actually *lost* 1.7% and 6.5% of their body weight within the first 7 exposure days, gaining weight at control or near control levels after that time. Early weight losses were also observed at 60 and 120 ppm among pregnant rats and rabbits in the developmental toxicity study of John (1983). In view of the studies showing actual body weight losses at concentrations close to the 49 ppm BMCL, the latter value was considered appropriate to evaluate acute and short-term risk.

In addition to the uncertainty associated with using body weight decrements as toxicologically significant endpoints, uncertainty was attached to the process of estimating the human equivalent concentrations (HECs) used to calculate MOEs. For the acute / short-term assessment, the body weight decrements were assumed to result from systemic exposure, in recognition of the fact that inhaled 1,3-D is indeed absorbed (Waechter *et al.,* 1992) (Stott and Kastl, 1986) and has toxicologic impacts distal to the lung in both rat and mouse, particularly on the bladder (Stott *et al.,* 1984). In the absence of specific information on the blood / vapor distribution of inhaled 1,3-D in both experimental animals and humans, we adopted USEPA's convention of defaulting the systemic RGDR scalar to 1 (USEPA, 1994), thus adjusting the BMCL only for the experimental animal and human exposure times and for the purity of the test article. According to the USEPA document (p. 4-61), "analysis of the available data on rats for blood:air partition coefficients shows that the $(H_{b/g})_A$ is greater than $(H_{b/g})_H$ in most cases". In that light, the default RGDR of 1 is viewed as a conservative estimate, though it should be mentioned that USEPA made the same assumption in its own risk assessment of 1,3-D (USEPA, 2007).

Furthermore, it was at least plausible that the body weight effect was NOT systemic in nature, but rather resulted from portal-of-entry impacts on the nasal passages and lung. While there were no experimental data to support this contention, longer-term exposures resulted in nasal and lung pathology, the indicators used to calculate seasonal, annual and lifetime (oncogenic) risks. There was precedent for a predominantly respiratory system impact leading to body weight decrements. Fischer 344 rats exposed to gaseous acrolein, a closely related chemical, exhibited body weight gain decrements without clear systemic toxicity at the doses employed (Dorman *et al.*, 2008). Upon removal from the daily exposure regimen at 13 weeks, body weights immediately began to correct toward control values, suggesting respiratory irritation as the basis for the effect. In another study, tracheal instillation of hydrochloric acid in C57BL6 mice resulted in body weight loss accompanied by several indicators of lung injury including decreased oxygenation, increased respiratory elastance, pulmonary inflammation, alveolar-capillary barrier dysfunction and epithelial injury (Patel *et al.*, 2012). Here too, the body weight effect probably stemmed from the initial respiratory tree impact. Calculation of the HEC for a portal-of-entry mediated effect could have invoked a rat-to-human whole-lung RGDR of 2.91 (calculated for male rats), thus raising the HEC and MOE by that factor. Or if an extrathoracic impact was sufficient to impact body weight, the RGDR would have been closer to 0.1. The default systemic RGDR of 1 obviously fell between these two possibilities.

Due to the use of the RGDR approach, the conventional interspecies uncertainty factor of 10 was reduced to 3, as RGDR is considered to remove the 3-fold pharmacokinetic portion while retaining the 3-fold pharmacodynamic portion of the interspecies factor (USEPA, 1994). The 10-fold intraspecies factor was retained, as there was no indication of the range of sensitivity at any exposure length within the human population. In fact, the only experimental evidence for varied sensitivity was provided by Rick (1988), who determined the odor threshold for Telone II among

22 adults to range between 1.8±1.2 ppm through 16.0±1.5 ppm, reasonably close to the default 10-fold intraspecies factor used in this analysis.

Despite the lack of evidence for developmental or reproductive toxicity, no data were available for any exposure length to assess the possibility of special inhalation sensitivity of infants or adolescents. Consequently, an additional database uncertainty factor of 3 was designated to protect these populations under scenarios where exposure might be anticipated. Three was chosen over 10 in recognition of the relative mildness of the critical endpoints for acute, subchronic and chronic toxicity. Nonetheless, the uncertainties inherent in the choice of such a factor are recognized.

2. Subchronic / seasonal risk

Subchronic / seasonal risk was evaluated using a critical $BMCL_{10}$ of 16 ppm based on very slight nasal hyperplasia in 2/10 male rats at 30 ppm after 13 weeks of daily exposure (5 days/week, 6 hr/day), which rose to 10/10 males and 10/10 females at 90 ppm (Stott *et al.*, 1984). The mildness of the sign combined with its appearance in a distinct minority of animals and with its apparently reversibility in rats (interim histopathology in the 2-yr rat inhalation study conducted at 6 and 12 months did not evidence this sign (Lomax *et al.*, 1987)) suggested that it might not be sufficiently adverse to drive the seasonal risk evaluation. However, the fact that incidence reached 100% at 90 ppm, and severity had increased from "very slight" to "slight" by 150 ppm, suggested that it was indeed treatment-related and potentially adverse even at 30 ppm.

Uncertainty accompanied the use of RGDR-based portal-of-entry dosimetry to estimate seasonal HECs. First, the default input parameters for minute volume and extrathoracic surface area in rats and humans used to calculate extrathoracic RGDRs obscured the intra-individual variability in these essentially morphometric parameters. Second, use of RGDR did not take into account the possibility that tissue level---as opposed to organ level---differences likely exist between species. Consequently, only the default pharmacokinetic uncertainty factor of 3 was avoided, while the pharmacodynamic (*i.e.*, receptor level) uncertainty factor of 3 was retained. And third, as explained in a recent USEPA position paper, the validity of this approach to portal-of-entry dosimetry in the extrathoracic region was contingent on assumptions of uniform air flow, gas / vapor deposition and tissue surface areas throughout the extrathoracic area, none of which may be operative in the current case (USEPA, 2012a). In essence, normalization of minute volume to extrathoracic surface area as recommended by USEPA in its 1994 position paper (USEPA, 1994)---and as done for this analysis---carried considerable uncertainty. That approach yielded RGDRs that were notably less than one both for rats and mice, meaning that a proportionately lower amount of inspired air is required in humans to create an exposure equivalent to rats, with resultant HECs that are also lower than the rodent exposure concentrations. USEPA's 2012 account maintains that recent pharmacokinetic and computational fluid dynamic models set the extrathoracic dose adjustment factor (equivalent to the RGDR) at ≥1 for many chemicals. However, absent both 1,3-D specific data and the time to analyze USEPA's position in greater depth, we have chosen to retain the default RGDR default approach, described above, which

compares minute volumes to extrathoracic surface areas in rodents and humans, while recognizing that the subchronic and chronic HECs may be low estimates.

3. Chronic / annual (non-oncogenic) risk

Chronic / annual (non-oncogenic) risk was evaluated using a critical $BMDL_{10}$ of 6 ppm. This was based on dose dependent nasal histopathologic signs emerging in the 2-yr mouse inhalation study of Stott (1987) at the LOEL of 20 ppm. The signs included hyperplasia and hypertrophy of the urinary bladder transitional epithelium and hyperplasia of the nasal respiratory epithelium in both males and females, though females were more sensitive. Because the respiratory signs occurred at similar doses to those of the urinary bladder, we decided to base the risk evaluation on the former, as this entailed the use of the extrathoracic RGDR of 0.198 rather than a default systemic RGDR of 1. The respiratory based point of departure thus protects against the bladder pathology, which presumably resulted from systemic exposure. Even so, we recognize that in the chronic case as well as the subchronic (see previous section) the RGDR may turn out to be close to 1 even for extrathoracic effects.

4. Oncogenic risk

1,3-D was oncogenic in several systems. Most relevant to the current assessment, inhalation exposure induced statistically significant pulmonary bronchioloalveolar adenomas in male mice, most of which were detected at study termination following 2 years of exposure---5 days/wk, 6 hr/day---to 60 ppm. Incidence may have been elevated at 20 ppm, as well, though pairwise statistical significance was not attained, nor was that rate above historical control levels (see discussion, section IV.A.3.c.). Potency was assessed under the assumption that 1,3-D acted directly on respiratory tissue at the portal of entry, not requiring absorption or distribution to exert its effect.

Support for 1,3-D-induced carcinogenesis was forthcoming from several other laboratory animal studies. Stott (1995) showed that dietary administration of 1,3-D for 2 years produced benign liver adenomas in rats of both sexes (liver is a portal of entry for oral exposure) at a daily dose of 25 mg/kg/day, with probable induction in males at 12.5 mg/kg/day. Van Duuren (1979) demonstrated a statistically significant elevation of local fibrosarcomas (6/30 animals) following weekly subcutaneous injection of *cis*-1,3-D in mice for up to 2 years at 3 mg/injection. These investigators also observed non-statistically significant elevations of skin papillomas following (a) a single dermal application of *cis*-1,3-D (122 mg/mouse) followed by 3x/wk applications of 5 µg phorbol myristate acetate for at least 257 days (4/30 mice); and (b) 3x/wk dermal applications of *cis*-1,3-D (122 mg/mouse) for up to 594 days (3/30 mice).

Oncogenesis was also evident in NTP's lifetime gavage study in rats and mice (NTP, 1985). In rats exposed 3x/week for up to 27 months to 25 and 50 mg/kg/day, 1,3-D induced forestomach squamous cell papillomas and squamous cell carcinomas in males (50 mg/kg/day), as well as hepatic neoplastic nodules in males (25 and 50 mg/kg/day). Exposed females may also have exhibited increased squamous cell papillomas at both doses, though the data were less clear. In

mice exposed 3x/week to 0, 50 and 100 mg/kg/day for 104 weeks, neoplasia was evident in urinary bladder (transitional cell carcinomas: males at the high dose, females at both doses), lung (bronchioloalveolar adenomas: males and females at both doses; bronchioloalveolar carcinomas: males at both doses), and forestomach (squamous cell papillomas: males and possibly females at both doses; squamous cell carcinomas: high dose in females only). Epichlorhydrin, which has oncogenic properties of its own, acted as a stabilizer in the 1,3-D preparation used in the NTP study. However, it is unlikely that it was present in sufficient quantities to be responsible for the observations summarized here (Konishi *et al.*, 1980).

Finally, 1,3-D was implicated in human oncogenesis in two studies. The first, by Markovitz (1984), reported two fatal cases of histiocytic lymphoma that developed exactly 6 years after emergency responders were exposed to 1,3-D after a tank truck spill. Nine responders, including those that developed lymphoma, exhibited acute signs directly after the spill. Both lymphoma cases died the year after detection. The authors drew structural parallels between 1,3-D and two known human carcinogens: vinyl chloride and dibromochloropropene. In a second study, Clary (2003) used California's Pesticide Use Report to examine possible epidemiologic correlations between deaths from pancreatic cancer, 1989-1996, and exposure to organochlorine pesticides in Fresno, Kern and Tulare Counties, 1972-1989. Long-term residents (>20 years) showed an odds ratio of 1.89 for 1,3-D and pancreatic cancer mortality, the highest of any of the 15 compounds examined (captafol, pentachloronitrobenzene and dieldrin also showed correlations).

While there is little uncertainty regarding 1,3-D's ability to induce tumors in a variety of tissues, species and exposure routes, great uncertainty is attached to the estimation of oncogenic risk, particularly as that estimation is contingent on whether oncogenesis occurred via local (portal of entry) or systemic action. The supporting evidence for each of these routes was presented above in section IV.A.3.c. An assumption of portal of entry action resulted in air unit risk (equivalent to oncogenic potency) values that were 3.44 times *less* than the equivalent values evolving from an assumption of systemic action due to the effect of the RGDR scalar (3.44 for portal of entry, 1 for systemic). For this reason, oncogenic risk is presented as a range for each exposure scenario between the value calculated for portal of entry and that calculated for systemic action.

In the absence of confounding evidence, demonstration of genotoxicity is considered sufficient to invoke the multistage cancer model. Multistage modeling constrains the dose-response curve to linearity, such that even small concentrations of an oncogen are associated with increased cancer risk. In view of the evidence for 1,3-D's genotoxicity, we used this approach in the current assessment. The emergent oncogenic risk values---*i.e.*, the products of the air unit risk or cancer potency (*i.e.*, the slope of the air concentration or internal dose *vs.* tumor incidence relation) and the lifetime daily concentration, for bronchioloalveolar adenomas---were over 10^{-4} for some occupational scenarios and over 10^{-2} for tarp removers using this approach (Table IV.19).

However, there are reasons to question the multistage linear extrapolation approach for inhaled 1,3-D-induced lung tumors. Most importantly, the incidence curve for bronchioloalveolar adenomas---9/49, 6/50, 13/49 and 22/50 at 0, 5, 20 and 60 ppm---suggests the existence of an *effective* threshold for tumor production. In this view, very low concentrations of 1,3-D would *not* induce tumors since the organism has the presumed capacity to detoxify the chemical through metabolism and/or excretion.

A threshold-dependent non-linear mode of action would be supported by evidence that (1) 1,3-D is *not* genotoxic under physiological conditions, and (2) 1,3-D acts by promoting the expansion of previously initiated cells. With respect to the first point, several recent genotoxicity studies conducted with formulations that were either unstabilized or stabilized by epoxidized soybean oil instead of the mutagenic compound, epichlorohydrin, have proven negative. These include the mouse micronucleus study (Dow Chemical Company, 1985b), the "Big Blue" mouse *in vivo* inhalation study (Gollapudi and Cieszlak, 1997), the DNA adduct study in rat liver (Stott *et al.*, 1997) and mouse lung and the rat dominant lethal study (Gollapudi *et al.*, 1998). In addition, Creedy (1984) and Stott (2001) have maintained that physiological levels of glutathione should be adequate reduce or abolish latent genotoxicity induced by 1,3-D or its metabolites. However, there is ample evidence both from *in vitro* and *in vivo* testing to suggest that 1,3-D is in fact genotoxic. These include positive indications in Ames-*Salmonella* testing, mouse lymphoma cells, and in inducing chromosomal aberrations in bone marrow and micronucleated polychromatic erythrocytes upon intraperitoneal injection into mice. In addition, several of the prominent 1,3-D metabolites are Ames positive. These compounds include 1,3-D's epoxidated derivative, 3-chloro-2-hydroxypropanal and 3-chloroacrolein.

With respect to the second point, the evidence that 1,3-D is a promoter comes primarily from a study of 1,3-D induced GSTP negative foci in the livers of diethylnitrosamine (DEN) treated rats (Klaunig *et al.*, 2015). The major findings of that study were (1) daily gavage with 1,3-D for 30 or 60 days at the hepato-oncogenic dose of 25 mg/kg/day increased the number and size of such foci, and (2) both focus numbers and proportion of liver volume occupied for by foci returned to control levels during a 30-day recovery period. The implication of the Klaunig study is that the induced lesions were preneoplastic. Farber's group has described the neoplastic potential of similar precursor lesions in the rat liver (Farber, 1973; Solt *et al.*, 1977).

Several recent reviews on the general issue of the absence or presence of thresholds in chemical carcinogenesis emphasized the difficulty of convincingly establishing thresholds in oncogenesis, especially considering the complications of hormetic effects, metabolism and the statistical power inherent in standard study designs (Purchase and Auton, 1995); (Fukushima *et al.*, 2005); (Neuman, 2009). It may also be relevant that most oncogens have both initiating and promoting capabilities. In this light, two caveats should be recognized before applying Klaunig's

observations to 1,3-D induced pulmonary oncogenesis. First, those observations were made in the rat liver, not the mouse lung, where exposure to 1,3-D vapor was shown by Stott *et al.* to produce adenomas (Stott *et al.*, 1987). Moreover, inhaled 1,3-D did not induce adenomas in rat lung (Lomax *et al.*, 1987), suggesting---if indeed this mode of action was a necessary precursor to tumor formation in response to 1,3-D---that rat lung had little to no ability to form preneoplastic foci. And second, Klaunig *et al.* did not demonstrate in the course of their 1- or 2-month oral exposures any progression from induced foci to actual tumors (without a doubt, the 1-3 month duration of the study was insufficient time for this to occur). While Farber's group indeed showed progression from preneoplastic foci to tumors in DEN-initiated rat livers after subsequent co-treatment with acetyl aminofluorine (to inhibit normal liver cell multiplication) and partial hepatectomy (to stimulate the growth of initiated cells), such a demonstration with 1,3-D would be necessary in order to carry the point that 1,3-D acts as a tumor promoter even in the liver, not to mention the lung. Finally, it should be noted that there was absolutely no evidence for preneoplastic foci in the mouse lung after 2 years of inhalation exposure to 1,3-D, despite the appearance of adenomas.

For the reasons discussed above, we considered multistage modeling, with its low-dose linear constraints, to be the most appropriate approach to evaluating the oncogenic risk of 1,3-D. The resultant risk values for many non-occupational and occupational scenarios, expressed as the probability of cancer in humans exposed under specified conditions, were above the negligible risk standard of 10^{-6}. Nonetheless, a further uncertainty regarding mode of action exists. If it emerged that the oncogenic action of 1,3-D was entirely as a non-genotoxic promoter that operated with a threshold, then a tumor NOEL and resultant MOEsmay be the more appropriate risk metric. However, since a threshold mechanism was not identified for this compound, and since insufficient animals were available to establish a threshold, oncogenic MOEs were not calculated.

5. Uncertainties due to metabolites, degradates, impurities and co-formulated chemicals

Metabolism or degradation of 1,3-D produces compounds that are chemically reactive and potentially toxic, genotoxic or oncogenic. These include the chloroallyl alcohols, chloroacrylic acids and chloroacroleins. While the toxicity of these compounds is no doubt modulated by glutathione conjugation to produce excretable mercapturic acid derivatives (Stott and Gollapudi, 2001), their concentration and duration in target organs such as the lung is essentially unknown. 1,3-D formulations contain similar compounds---or will produce them on storage. This analysis did not assess the risk of metabolites and degradates, assuming instead that their appearance under the conditions present in the toxicity studies would be accounted for in the effects generated. Even so, an understanding of 1,3-D's toxicity is not complete without also understanding the toxic properties of metabolites, degradates and impurities, particularly as conditions in the field may conceivably affect the relative concentrations.

Finally, it is worth mentioning that most 1,3-D-containing formulations sold in California also contain the fumigant chloropicrin, which is not only a severe irritant, but also may be carcinogenic. That the two chemicals could have synergistic toxicity or carcinogenicity is not known, but is considered at least a plausibility.

B. EXPOSURE APPRAISAL

In this exposure assessment, the occupational and occupational bystander exposure estimates were updated using the AGRIAN® PUR records specific to 1,3-D. As detailed in Section C, PRODUCT FORMULATIONS AND USES, the AGRIAN® database contains more detailed and up-to-date information on 1,3-D use (*e.g.*, specific method of application, application date, application company, application rate) than the DPR-PUR database. SAC, AAC, and LAC exposure estimates generated using the AGRIAN® PUR records are more accurate than those generated using the DPR-PUR database. The increased amount of use data allows for estimation of application method-specific use seasons and seasonal application rates. Moreover, for each application method, these estimates were generated for the company applying the most 1,3-D in the highest use county for that method. Had the DPR-PUR database been used for estimating the SAC, AAC, and LAC exposure estimates, a single use season and estimated seasonal application rate would have to have been used for all of the exposure scenarios.

1. Occupational Exposure Estimate Uncertainties

a. Applicator (Shallow Shank with Tarp, Drip with Tarp, Drip without Tarp, Deep Shank with Tarp, Injection Auger, and Tarp Remover)

The exposure estimates generated for handlers are based upon certain assumptions. The use of surrogate data from studies monitoring chloropicrin to estimate 1,3-D air concentrations for these scenarios relies upon two assumptions. The first assumption is that 1,3-D breathing-zone air concentration ratios would mimic those of chloropicrin. However, that assumption may be false. Another potentially incorrect assumption is that spillage controls were not used for the shank injection portion of the chloropicrin study. With the exception of the shallow shank applicator using a tarp, or deep shank applicator using a tarp, this assumption led to the use of the corresponding 1,3-D data for estimating breathing-zone air concentrations.

The injection auger applicator exposure estimates rely upon assumptions concerning the maximum application rate. The maximum application rate used for the short-term air concentration estimate is based on chloropicrin surrogate data, and the assumption that all 248 tree replant sites would be located on one acre. This approach may not provide an accurate maximum of the injection auger application rate for 1,3-D was but was used because data were not available.

The estimated breathing-zone 1,3-D air concentrations for the tarp remover are not representative of this handler using respiratory protection. Due to the previously described lack of respiratory requirements for this scenario on some of the labels for active products on DPR’s product label list, respiratory protection was not incorporated into the calculated estimates. Hence, these estimates are anticipated to be representative of the 1,3-D air concentrations inhaled by the tarp remover not wearing respiratory protection. However, they are likely higher than those for the tarp remover utilizing respiratory protection.

There's increased uncertainty in the estimated breathing-zone 1,3-D air concentrations for handlers injecting Tri-Cal Trilone II. The label for this active product lacks language, present on the other applicable labels, prohibiting the substitution of an enclosed tractor cab, equipped with filtration, for a respirator. A protection factor for a half-face respirator was incorporated into the exposure estimate calculations, reducing estimated breathing-zone 1,3-D concentrations. However, an enclosed tractor cab may not provide as much protection as the respirator, leading to breathing-zone 1,3-D air concentrations higher than those anticipated.

b. Reentry worker

Use of the reentry worker exposure study data conducted by the registrant may have led to estimates higher than the actual exposure for this worker. The workers in the study reentered the treated field well before the end of the current CA 7-day REI. The exposure data for the worker reentering the field after about 3.8 days, the longest interval of the study, was used to estimate exposure for this scenario.

c. Occupational bystander

The estimated breathing-zone air concentrations for the occupational bystander were based upon simulated and ambient measured air concentrations. However, the simulated air concentrations for shank-injection without the use of a tarp may not accurately represent occupational bystander exposures from shank injections where tarps are applied. Likewise, the simulated air concentrations for drip chemigation with the use of a soil-sealing tarp may not be representative of air concentrations for drip chemigation without the use of a tarp: one of the products containing 1,3-D allows for drip-chemigation without the use of a tarp, albeit at a reduced application rate.

As mentioned earlier, the SAC, AAC, and LAC values were generated using the 1,3-D air concentrations measured in Merced County during the estimated use seasons (2 use seasons of 4 months each), in that county for 2011. However, to avoid using data which exceeded the annual township cap, only a portion (i.e., 6 of the 8 months), of the total use season data was used to estimate exposure. Each of these months was shown to have a lower amount of 1,3-D applied than the corresponding month in Fresno County, a higher use county, in 2011. However, using 6 months vs. 8 months of the seasonal data to estimate the SAC may provide less accurate estimates of the SAC, AAC, and LAC. Moreover, Merced County was not the highest use county in 2011. These issues may lead to underestimation of exposure. However, due to the lack of data this approach was taken.

d. Comparison of the Occupational Exposure Estimate Air Concentrations generated in EPA's 2007 Risk Assessment Document (RAD) and the Current EAD

EPA assessed only the applicator (shallow shank w/o tarp), loader, reentry worker, and occupational bystander exposure scenarios. Hence, the breathing-zone air concentration estimates generated by EPA and DPR will be compared for just these scenarios. The label used in EPA's RAD for the air concentrations for these scenarios is "□g/m^3". This label was

interpreted as being "µg/m^3". In order to compare the EPA values with those of this RCD, the EPA values were converted to ppm.

Both EPA and DPR utilized the Houtman study for estimating exposures (Houtman, 1993). However, EPA used additional registrant worker exposure studies for minibulk loaders and applicators (MRID 43880401) and for worker exposures associated with Yetter rig fumigation (MRID 45120702). Neither of these latter two studies was used to assess exposure in the current EAD. The samples obtained in the first of these studies were task-specific samples which are relatively short (≤ 46 minutes), and too biased towards a specific high-exposure potential task for estimating representative 8-hr TWA exposure estimates. The second of the studies utilized the Yetter application rig which is not currently used in CA.

The 1,3-D air concentrations derived from the registrant studies in EPA's RAD for estimating applicator (shallow shank w/o tarp), loader, and reentry worker exposures differ from those in the current EAD. For the short-term exposures, EPA derived an air concentration of 1.4 ppm for the applicator (shallow shank w/o tarp) while DPR calculated an air concentration of 0.27 ppm. EPA's applicator (shank) air concentration is higher than that used for estimating the STAC in the current EAD because EPA included task specific samples (≤ 46 minutes) from the registrant study. These air concentrations were generally higher than the 4-hr TWA samples used to estimate exposure in the current EAD since they were only taken during high exposure potential activities. For the current EAD, only 4-hr TWA samples were utilized since they better represent an 8-hr TWA exposure. Moreover, EPA did not adjust the breathing-zone air concentration for respiratory protection. A half-face respirator would reduce the estimated air concentration in the breathing-zone by 10-fold (i.e., 0.14 ppm). EPA's loader short-term air concentration of 7.1 ppm is substantially higher than that used for estimating the STAC in the current EAD because EPA used only task specific samples (≤ 46 minutes) from the registrant study. As with the applicator (shank) portion of the study, these air concentrations were generally higher than the 4-hr TWA samples used to estimate exposure in the current EAD since they were only taken during high exposure potential activities. In addition, the loader air concentration from EPA's RAD is higher since a respiratory protection factor was not incorporated into the result. A half-face respirator would reduce the air concentration in the breathing-zone by 10-fold (i.e., 0.71 ppm). The reentry worker air concentration used to estimate exposure in the EAD is lower than that in the RAD because only the air concentration data from the study with the longest reentry interval were used. Even so, this interval (~3.8 days), is only about half as long as the CA REI of 7 days. Because air concentrations decrease with additional time post-injection, it likely overestimates exposure. Respiratory protection was not factored into the EAD air concentration since, according to product labels and permit conditions, respiratory protection is not required for reentry workers (USEPA, 2007) (Table V.1).

Exposure estimate air concentration differences between EPA's RAD and the current EAD also exist for the occupational bystander scenarios. Both organizations used ISCST3 modeling to derive the breathing-zone air concentrations for these scenarios. However, the air concentrations

were modeled for different sized treated fields and at different distances from the field-edge. For shank injection, EPA utilized a field-size of 40 acres for the simulation while DPR used 80 acres. In addition, air concentrations were simulated much closer to the field-edge by DPR (3.04 meters) than EPA (25 meters). For drip chemigation, both EPA and DPR utilized a field-size of 40 acres for the simulation. However, as with shank fumigation, the air concentrations were simulated much closer to the field-edge by DPR (3.04 meters) than EPA (25 meters) (U.S. EPA, 2007) (Table V.3). The DPR simulated air concentrations for shank and drip-chemigation presented in Table V.3 are the highest values generated in the DPR simulation study for these application methods (Johnson, 2009a).

Table V. 1 Comparison of EPA 2007 Risk Assessment Document (RAD) and Exposure Assessment Document (EAD) Breathing-Zone Air Concentration Estimates (ppm)

Scenario	RAD		EAD	
	Short-term [a]	Long-term [b]	Short-term [c]	Long-term [d]
Applicator (shank) [e]	1.4	0.03	0.52	0.04
Loader [f]	7.1	0.11	0.45	0.06
Reentry Worker [g]	0.22		0.04	
Occupational Bystander (shank) [h]	1.1		2.0	
Occupational Bystander (drip) [i]	0.54		0.86	

[a] Air concentrations were used to directly generate acute Margins of Exposure (MOE's) in 2007 EPA RAD (USEPA, 2007). No absorbed dosages were calculated.

[b] Air concentrations were used to estimate the lifetime average daily exposures (LADE) in 2007 EPA RAD (USEPA, 2007). The air concentration listed in the RAD for calculating the LADE are median values and were reduced by 10X since the LADE values incorporated respiratory protection. No absorbed dosages were calculated.

[c] 95th %-ile of air concentrations which have been adjusted for the maximum application rate and for respiratory protection, if applicable, and then subsequently used to calculate the Short Term Air Concentration (STAC) values for the current EAD.

[d] Mean of air concentrations which have been adjusted for the seasonal application rate and for respiratory protection, if applicable, and then subsequently used to calculate the Seasonal Air Concentration (SAC) which is then used to calculate the Annual Air Concentration (AAC) which is used to calculate the Lifetime Air Concentration (LAC) values for the current EAD.

[e] EPA's short-term applicator (shank) air concentration is higher than that used for estimating the STAC in the current EAD because EPA included task specific samples (≤ 46 minutes) from the registrant study (USEPA, 2007). These air concentrations were generally higher than the 4-hr TWA samples used to estimate exposure in the current EAD since they were only taken during high exposure potential activities. For the current EAD, only 4-hr TWA samples were utilized since they better represent an 8-hr TWA exposure. Moreover, EPA did not adjust the breathing-zone air concentration for respiratory protection. A half-face respirator would reduce the air concentration in the breathing-zone by 10-fold (i.e., 0.14 ppm).

[f] EPA's short-term exposure loader air concentrations are substantially higher than that used for estimating the STAC in the current EAD because EPA used only task specific samples (≤ 46 minutes) from the registrant study. These air concentrations were generally higher than the 4-hr TWA samples used to estimate exposure in the current EAD since they were only taken during high exposure potential activities. In addition, the loader short-term air

concentration from EPA's RAD is higher since a respiratory protection factor was not incorporated into the result (USEPA, 2007). A half-face respirator would reduce the air concentration in the breathing-zone by 10-fold (i.e., 0.71 ppm).

[g] The reentry worker air concentration used in the "EAD" column is lower because only the air concentration data from the study with the longest reentry interval were used. Even so, this interval (~3.8 days), is substantially less than the CA restricted entry interval (REI) interval of 7 days. Hence, it likely leads to an overestimation of exposure. Respiratory protection was not factored into the air concentration since, according to product labels and permit conditions, respiratory protection is not required for reentry workers.

[h] RAD: ISCST3 model simulation of air concentration for maximum application rate at 25 meters from edge of 40-acre field undergoing shank fumigation (8-hr exposure) (USEPA, 2007). EAD: ISCST3 model simulation of air concentration for maximum application rate at 3.04 meters from edge of 80-acre field undergoing shank fumigation (8-hr exposure) (Johnson, 2009a).

[i] RAD: ISCST3 model simulation of air concentration for maximum application rate at 25 meters from edge of 40-acre field undergoing drip fumigation (8-hr exposure) (USEPA, 2007). EAD: ISCST3 model simulation of air concentration for maximum application rate at 3.04 meters from edge of 40-acre field undergoing drip fumigation (8-hr exposure) (Johnson, 2009a).

2. Residential Bystander Exposure Estimate Uncertainties

All residential bystander exposures discussed in this document are based on the assumption that a resident will spend 24 continuous hours either at 100 feet from a treated field, or in an area with elevated ambient air concentration of 1,3-D, or both. This assumption may lead to an overestimation of human exposure.

a. Residential Bystander Exposures Associated with Nearby Field Applications

The majority of 1,3-D product labels and CA permit conditions for products containing chloropicrin, mandate a 100-foot buffer zone between the fumigated field and occupied structures. All but three labels (Telone EC, Telone II and TriCal Trilone II) also require that all non-handlers, including field workers, residents, pedestrians, and other bystanders, must be excluded from the buffer zone during the buffer zone period. These 3 labels allow the possibility that residents spending time outdoors are within the limits of the buffer zone. These residents would experience 1,3-D exposure higher than the residential bystander exposures presented in this document for the edge of the buffer zone. Although we expect such exposures to be rare and short in duration (up to several hours), the possibility of accidental acute bystander exposures of this kind remains. These exposures will approximate the occupational bystander exposures outlined in Table IV.8.(Occupational Exposure Estimates) and will be in the range of 0.6-2.0 ppm for an 8-hr exposure, depending on the application method.

b. Residential Bystander Exposures Associated with Tree and Vine Applications

The short-term residential bystander exposures summarized in Table IV.8 were estimated using air concentrations modeled at 100 feet from a field treated with 344 lb 1,3-D per acre. A slight overestimation of the exposure scenarios listed in Table IV.8 comes from the use of this application rate in the air dispersion modeling. The California Permit Conditions cap the

application rate of 1,3-D to 332 lb AI/acre (see discussion in Regulatory Status section of this document).

The following 1,3-D products do not contain chloropicrin: Telone EC, Telone II, and Tri-Cal Trilone II. Chloropicrin is not required as a warning agent in 1,3-D soil fumigations. As such, these products do not fall under the buffer zone restrictions of the California permit conditions for chloropicrin. In fact, the labels for these three products do not require buffer zones if the product "will be used on soils that would not experience an additional 1,3-D treatment for at least three years, for example, on soils to be planted with perennial crops. For example, on soils to be planted with fruit trees, nut and nursery crops, perennial vines, hops, mint or pineapple". The exposures presented in Table IV.8 may be underestimated under the situations in which Telone EC, Telone II, or Tri-Cal Trilone II are used for soil fumigations and buffer zones are not established around the treated fields.

The tree and vine exposure data were modeled assuming a maximum of 74.4 acres treated. To determine if this maximum is realistic, the California pesticide use database (PUR) was searched for daily tree and vine applications in 2009. Of the 481 records retrieved, all but 8 were 74.4 acres or less. The eight that exceeded this maximum ranged in size from 75.3 to 229 acres. It is important to recognize that sometimes multi-day applications are reported as a single day. However, the assumed application size could underestimate some applications. For acute exposures, it is expected that some actual exposures could be equal to or greater than the values listed in Table IV.8.

c. Residential Bystander Exposures Associated with Ambient Air

The Parlier study in 2006 showed that some 1,3-D ambient air concentrations may exhibit a potential of health concern (Wofford *et al.*, 2009). This finding prompted DPR to request Dow AgroSciences to conduct an additional study in 2010-2012 (Rotondaro and Van Wesenbeeck, 2012a). The ambient air monitoring study in Merced County showed even more elevated ambient air concentrations, with the highest short-term-air concentration exceeding the previous records by orders of magnitude. It is possible that the 24-hr 1,3-D ambient air concentrations in high 1,3-D use areas are even higher that those recorded in both Parlier and Merced studies. One reason is the method of air sampling in both studies. Only a limited number of samplers were employed: a single receptor in Parlier, and nine receptors in Mercer County. Collecting 72-hr samples in the Merced study most likely missed 24-hr concentration peaks of 1,3-D. In the Parlier study, 24-hr air samples were collected every three days during the high 1,3-D use season, and it is possible that 24-hr concentrations higher than 23.6 $\mu g/m^3$ occurred on the two days without air monitoring. Additionally, the highest three-day air concentration of 1,3-D in Merced County was recorded on December 14th, 2011, at Township #5, and it was 369.2 $\mu g/m^3$ or 0.0813 ppm. The sample preceding it (from December 11th, 2011) was missing due to equipment theft from the field, and the air monitor was moved ¼ mile away on December 11th. These factors introduce uncertainty in the estimates of the short-term residential bystander exposures to ambient air.

Both Merced County and Fresno County were receiving a high number of township cap exceptions in the years when the Parlier (2006) and the Merced (2010-2012) studies were conducted. The township cap in California is set at 90,250 lb per township annually. Five of the nine townships included in the DAS study received 1,3-D close to or exceeding the township cap (between 101,258 lb and 256,712 lb of 1,3-D were applied in the 5 townships during the 14.5 months of the study). The amount of 1,3-D applied in the Parlier area during the 12 months of monitoring was recorded only for the 6-7 miles around Parlier (302,075 lb, or 23% of the 1,305,054 lb AI used in Fresno County in 2006), but not beyond that area. The Merced study was selected for estimating bystander exposures to 1,3-D in ambient air on the grounds of the robustness and sensitivity outlined in the Exposure Assessment section. However, because of the exceedance of the township cap in the monitored areas during both studies, the short-term and seasonal exposure estimates presented in this risk assessment may overestimate the exposures in areas where the 1,3-D township cap is in compliance.

d. Combined Residential Bystander Exposures Associated with Nearby Field Applications and Ambient Air

For people residing in the high-use 1,3-D townships, it is possible to experience short- or long-term exposures to elevated 1,3-D ambient air concentrations while spending time in close vicinity to ongoing 1,3-D applications. The worst case scenario for combined bystander exposure at 100 feet from a shallow shank application (highest exposure potential scenario for a field application) and ambient air is presented in Table V.2. below. All data are from Table IV.8.

Table V. 2 Combined Residential Bystander Exposures Associated with Nearby Field Applications and Ambient Air (ppm)

Exposure scenario	STAC [a]	SAC [b]
Combined exposure from nearby shallow shank application and ambient air	0.6321	0.0218

[a] STAC: Short-Term Air Concentration: Sum of STAC estimated at 100 feet downwind from the edge of a field fumigated by shallow shank, and STAC for ambient air (0.5508 ppm + 0.0813 ppm = 0.6321 ppm) (Table IV.8).
[b] SAC: Seasonal Air Concentration: Sum of SAC estimated at 100 feet downwind from the edge of a field fumigated by shallow shank, and SAC for ambient air (0.0173 ppm + 0.0045 ppm = 0.0218 ppm) (Table IV.8).

e. Comparison of the Residential Bystander Exposure Estimate Air Concentration Generated in EPA's 2007 Risk Assessment Document (RAD) (USEPA, 2007) and the Current EAD

The two agencies use different definitions for short-, intermediate- and long-term exposures. EPA distinguishes between acute (less than 24 hours), short-term (1-30 days), intermediate- term (1 month- 6 months) or long- term (> 6 months) exposures (USEPA, 2007). For comparison, we define short-term exposure as exposure lasting from a day or less, and up to one week. Seasonal exposure is defined as a period of frequent exposure lasting more than a week but substantially

less than a year, whether the exposure is constant or intermittent during the period. Annual exposure integrates all exposure periods during the year. Lifetime exposures integrate all exposure periods over several years (Beauvais, 2006; Beauvais, 2012).

For near-field sources (farmfields), EPA assessed only acute (24 hours) non-occupational bystander exposure scenarios for shank and drip applications. EPA acknowledged that at the time the Risk Assessment Document was prepared (2007) the computer models could not readily be used for exposures of longer duration (USEPA, 2007). Hence, the air concentration estimates generated by EPA and DPR were compared only for short-term (24 hour) scenarios. As mentioned before, the EPA metric of the air concentrations for these scenarios was interpreted as "µg/m^3". Both EPA and DPR utilized ISCST3 modeling for estimating bystander exposures. However, the inputs for field and buffer zone sizes and for application rate in the models were different. For shank injection, the EPA simulations used 40 acres, 25 meters (82 feet) and 355 lb/acre while the DPR simulations utilized 80 acres, 100 feet and 332 lb/acre. For drip chemigation, both agencies utilized the same field size (40 acres) but the EPA simulations used 25 meters (82 feet) and 355 lb/acre, while DPR used 100 feet and 332 lb/acre. For the purpose of comparison, additional DPR short-term estimates for shank and drip applications were generated, in µg/m^3, and adjusted for the buffer distance (25 meters), acreage (40 acres), application rate (355 lb/acre) and atmospheric stability used by EPA. These new estimates were calculated using the same flux rate and air concentration data used to generate the estimates shown in Table IV.8 (Barry, 2015b). The estimates and the corresponding estimates from EPA are presented in Table V.3.

Table V. 3 Comparison of EPA 2007 Risk Assessment Document (RAD) and Exposure Assessment Document (EAD) Air Concentration Estimates (µg/m^3) for Residential Bystanders

Scenario	RAD		EAD	
	Short-term	Long-term	Short-term	Long-term
Shank injection	1970 [a]	N/D [b]	2429 [c]	
Drip irrigation	1510 [d]	N/D [b]	936 [e]	
Ambient air	128 [f]	13.30 [g]	369 [h]	0.99 [i]

[a] 24-hr air concentration, calculated using ISCST3 model, at 25 meters (82 ft) from a 40 acre field receiving shank injection at application rate 355 lb/a (Table X13 in (USEPA, 2007).
[b] Not determined.
[c] 24-hr air concentration, estimated using ISCST3 model, at 25 meters (82 ft) from a 40 acre field receiving shallow shank injection at application rate 355 lb/a for comparison to EPA data.
[d] 24-hr air concentration, calculated using ISCST3 model, at 25 meters (82 ft) from a 40 acre field receiving drip irrigation at application rate 355 lb/a (Table X12 in (USEPA, 2007)).
[e] 24-hr air concentration, estimated using ISCST3 model, at 25 meters (82 ft) from a 40 acre field receiving drip irrigation at application rate 355 lb/a for comparison to EPA data.
[f] 24-hr TWA air concentration, observed in Kern County in 2000 (Table 9 in (USEPA, 2007)). Converted from ppm to µg/m^3 (0.02825 ppm = 128 µg/m^3).

[g] 7 week mean air concentration, observed in Kern County in 2000 (Table 9 in (USEPA, 2007)). Converted from ppm to µg/m^3 (0.00293 ppm = 13.3 µg/m^3).
[h] 72-hour air concentration, observed in Merced county in 2011 (Rotondaro and Van Wesenbeeck, 2012a).
[i] Median of 129,600 simulated annual ambient air concentration values of Township #5 by SOFEA-2 (Table IV.8).

EPA also utilized PERFUM modeling for estimating bystander exposures to shank injection (USEPA, 2007). The PERFUM model used at the time the RAD was prepared allowed only buffer distributions, but not air concentrations in the output. This does not allow for direct comparison between DPR air concentrations and the EPA modeling.

For ambient air exposures, EPA and DPR utilized different studies. EPA used ARB studies conducted in California for 7-9 weeks during the "season of high use" (USEPA, 2007). Samples were collected for 24 hours, 1 to 4 times a week. The long-term ambient air concentration was calculated as the mean of weekly means for samples collected over the course of the calendar week. In contrast, DPR used a registrant study conducted in California for 14.5 months (Rotondaro and Van Wesenbeeck, 2012a). Continuous 72-hr samples were collected. The long-term ambient air concentration was calculated as the median of the annual average air concentrations simulated by the SOFEA model (Table IV.8). The highest measured short-term air concentration in Kern County in 2000 was lower compared to the respective air concentration in Merced County in 2011 (Table V.5). A possible reason for the difference in short-term air concentrations could be the increased use of 1,3-D above the township cap allocation in California after 2002. The highest long-term air concentration in Kern County in 2000 was higher than the long-term (annual) air concentration in Merced County in 2011. As stated earlier, the long-term air concentration used by EPA was generated from 7-9 weeks of air monitoring data obtained during the season of high use. If the same approach is used for analysis of the Merced study data, the mean 8-week ambient air concentration, recorded in the months of November and December 2011 in Merced County, would be approximately 40 µg/m^3.

g. Lifetime Exposure

Lifetime exposure estimates of residential bystanders are most applicable to the use pattern of 1,3-D that computer model employed. In this exposure assessment, air concentrations of 1,3-D were generated by SOFEA-2 based on the use pattern (i.e., use rates and crop specific uses) and weather conditions in Merced, CA, a high 1,3-D use area. Hence, it is reasonable to expect that these exposure estimates should be applicable to Merced and other areas with a similar or lower use level of 1,3-D. However, any significant increases in the use of 1,3-D that result in exceedance of the highest modeled annual average air concentration would require an update of the lifetime exposure estimates.

1,3-D Air Concentration

As mentioned previously, this risk assessment relied on a list of 100 average annual air concentrations from SOFEA-2 based on the pattern of 1,3-D applications in Merced, CA (Driver *et al.*, 2014a). Table V.4 shows the total number of simulated average annual air concentrations that is equal to or greater than the observed value in each of the 9 townships in Merced. The observed value was derived by averaging the 72-hour air concentrations in a particular township for 2011 from a monitoring study by the registrant (Rotondaro and van Wesenbeeck, 2012b). As can be seen in Table V.6, for each of the 9 townships, some stimulated values are higher than the observed value, indicating SOFEA was able to bracket the ambient air concentrations observed. However, for Township #2 and #5, <0.1% of the simulated values are greater than the observed. The infrequent occurrences of these high modeled concentrations in Township #2 and Township #5 may have resulted from some additional issues identified in the SOFEA-2: mixing-height correction and atmospheric stability class designation (Barry, 2015a). For assessing the lifetime exposure to 1,3-D, MCABLE employed the entire list of 100 average annual air concentrations for estimating exposures. Hence, the infrequent occurrence of high 1,3-D concentrations in the SOFEA predictions (especially for Township # 5) would likely result in under-prediction of the lifetime exposures by MCABLE.

Unlike MCABLE, HEE5CB employed a single list of average annual air concentrations for estimating the lifetime exposure to 1,3-D. To minimize the impact of infrequent occurrence of high 1,3-D air concentrations in SOFEA-2 predictions, for the HEE5CB simulations, the ranges of input air concentrations were restricted to those that bracketed the mean observed value in Township #5 (the highest measured 1,3-D concentrations were observed in Merced, CA). That is, only the simulation results with annual average values equal to or higher than the observed mean value of Township #5 were included. Accordingly, of the 100 lists of average annual air concentrations, 31 satisfied this criterion. Based on these lists of 31 average annual air concentrations, the LADD values from HEE5CB were presented in Table IV.11. As a reviewer pointed out, the selection criterion adopted may lead to over-prediction of the LADD and risk associated with 1,3-D exposure. However, since the inclusion of all data in MCABLE may result in some under-prediction, the “over-prediction of HEE5CB” and “under-prediction of MCABLE” may provide a range of realistic estimates of human exposures to 1,3 D in California.

Table V. 4 Numbers of SOFEA-2 Simulated Values Equal To or Greater Than the Annual Averages derived from the Ambient Measurement of 1,3-D at Merced, CA[a]

Merced Township	Mean (μg/m^3)[b] (Observed)	Simulated ≥Observed Mean (Number of Receptors)	Simulated ≥Observed Mean (Percentage)
1	0.83	1770	1.37
2	4.63	48	0.04
3	0.76	23936	18.47
4	1.39	2462	1.90
5	7.92	50	0.04
6	2.92	2344	1.81
7	0.27	129114	99.63
8	0.85	86998	67.13
9	0.51	122542	94.55

[a] A total of 129,600 simulated annual average air concentrations was generated by SOFEA-2 per township (i.e., 36 square mile/township x 36 receptors/square mile x 1 township = 1296 values per township; the simulation was repeated 100 times [i.e., 1296 x 100 =129,600 simulated values per township])

[b] Annual averages, as calculated in this risk assessment, were based on the 2011 monitoring results of 1,3-D by the registrant in Merced, CA (Rotondaro and van Wesenbeeck, 2012b).

Residency-Mobility

In MCABLE, the time of individuals spent within a high 1,3-D use area was derived from the results of a California-specific residential mobility survey (Kaplan, 2014) . In that survey, two high 1,3-D use areas were examined with Merced being the highest in terms of 1,3-D usages (Driver *et al.*, 2015). The survey protocol was reviewed and approved by DPR (Beauvais, 2013). Based on the information provided by the respondents from Merced, CA, a probability distribution of total time spent in their residence was estimated (Driver *et al.*, 2015). Additional adjustments to the probability distribution were made by these authors to account for the fact that (1) the survey focused on individuals of ≥18 years old (i.e., younger individuals were excluded; truncation error), (2) some respondents based (or anchored) their answers of the number of years stayed using a convenient value (e.g., a multiple value of 5 [i.e., anchoring bias]), and (3) unadjusted cross sectional survey data were considered insufficient (Powell, 2006) for use in estimating individual lifetime mobility (i.e., longevity bias). Using the adjusted probability distribution, the oncogenic risks of residential bystanders were estimated to be 2.87×10^{-6} in the males and 2.68×10^{-6} in the females. Because simulated individuals are "allowed" to move in and out of the high 1,3-D use area, intuitively, these oncogenic risk estimates would likely reflect the risk associated with the individuals who spent an "average" (or median) amount of time in the area. It is noteworthy that the 50th percentile value of the distribution of total time spent in Merced, CA, was estimated to be ~29 years (Driver *et al.*, 2015).

Similar to MCABLE, California-specific survey data (Wiley, 1991; Wiley *et al.*, 1991) were employed by HEE5CB for estimating the exposure within a high 1,3-D use area (Sanborn and Powell, 1994). However, comparing to the survey by Kaplen (2014), these survey data are generic (i.e., not region specific). Table V.5 shows a comparison of oncogenic risk between MCABLE and HEE5CB. Under the high mobility assumption of the HEE5CB, the 95th percentile oncogenic risk values associated with individuals lived from the first 30 years of their 70 year lifetime are $(3.12\text{-}4.85) \times 10^{-6}$ in the males and $(3.05\text{-}4.75) \times 10^{-6}$ in the females. The lower limits of these risk values based on the high mobility assumption are similar to those predicted by MCABLE. Given the fact that HEE5CB has more restrictive residency-mobility assumptions than MCABLE, this consistency in model outputs suggests that these models can provide a valuable insight into the range of exposures and oncogenic risks associated with the use of 1,3-D in California.

Table V. 5 Potential Lifetime Oncogenic Risk (95th percentile) Associated with 1,3-D Inhaled by Residential Bystanders in a High 1,3-D Use Area[a]

Residence Time	MCABLE	
	Male	Female
	95th Oncogenic Risk x 10^6	
Variable (Without Time-Away)	2.87	2.69
Residence Time	**HEE5CB**	
	Male	Female
	95th Oncogenic Risk x 10^6	
Birth to age 30 (High Mobility)[b]	3.12-4.85[c]	3.05-4.75[c]

[a] Risk was derived from the cancer risk distribution generated by MCABLE or HEE5CB. The individual risk value that constituted the cancer risk distribution was calculated as the LADD (μg/kg/day) multiple by the human cancer potency factor (portal-of-entry effect) of 0.000014 $(\mu g/kg/day)^{-1}$.

[b] Individuals are assumed living their first 30 years of their 70-year lifetime in the high 1.3-D use area. Exposure was estimated using the air concentration distributions from all the 3x3 townships.

[b] The lower and upper bound values represent the lowest and highest exposure estimates based on a list of 31 average annual air concentrations generated by SOFEA-2.

C. CRITICAL TOXICOLOGIC ENDPOINTS: USEPA *vs.* DPR

USEPA completed a reregistration eligibility decision (RED) on 1,3-D in 2007, establishing toxicologic endpoints for oral, dermal and inhalation exposures (USEPA, 2007). A revision followed in 2008 (USEPA, 2008a). Table V.6 details the inhalation endpoints as they appear in the 2008 update and compares them to the DPR endpoints established in the present document.

The large difference between the two agencies in the acute endpoint values reflects the Human Health Assessment Branch's decision---for the purposes of this assessment---to use short-term toxicologic values to represent acute exposure scenarios. With respect to the short-term scenario itself, the Human Health Assessment Branch opted to apply benchmark concentration modeling to the short-term body weight data, as shown above in Table IV.1. Other differences between USEPA and DPR-HHA reflect minor differences in the application of the RGDR scalar used to calculate human equivalent doses.

Table V. 6 Summary of 1,3-D inhalation endpoints used by USEPA (2008a) and the present assessment.

Exposure scenario	Animal endpoint values	Study & toxicologic effects (study type)	HECs & UFs
Acute			
USEPA (2008)	NOAEL=454 ppm LOAEL=583 ppm	Occupational Clinical signs, ↓BW, death ≥647 ppm (rat acute inhalation tox.)	Occupational HEC=227 ppm UF=30
		Non-occupational Clinical signs, ↓BW, death ≥647 ppm (rat acute inhalation tox.)	Non-occupational HEC=75.7 ppm UF=30
DPR-HHA (2015)	$BMCL_{1\sigma}$=49 ppm LOEL and NOEL n/a	Occupational ↓BW gain (rat subchronic inhalation toxicity)	Occupational HEC=33 ppm UF=30
		Non-occupational ↓BW gain (rat subchronic inhalation toxicity)	Non-occupational HEC=11 ppm UF=30 (100 for children)
Short-term (1-30 days)			
USEPA (2008)	NOAEL=20 ppm (maternal) LOAEL=60 ppm	Occupational ↓ maternal BW gain (rabbit dvp. toxicity)	Occupational HEC=15 ppm UF=30
		Non-occupational ↓ maternal BW gain (rabbit dvp. toxicity)	Non-occupational HEC=5 ppm UF=30
DPR-HHA (2015)	$BMCL_{1\sigma}$=49 ppm LOEL and NOEL n/a	Occupational Body wt. decrement (rat subchronic inhalation toxicity)	Occupational HEC=33 ppm UF=30
		Non-occupational ↓BW gain (rat subchronic inhalation toxicity)	Non-occupational HEC=11 ppm UF=30 (100 for children)

Intermediate-term (1-6 months)			
USEPA (2008)	NOAEL=10 ppm LOAEL=30 ppm	Occupational Nasal histopathology (rat 13-wk inhalation tox.) Non-occupational Nasal histopathology (rat 13-wk inhalation tox.)	Occupational HEC=0.86 ppm UF=30 Non-occupational HEC=0.205 ppm UF=30
DPR-HHA (2015)	$BMCL_{10}$=16 ppm LOAEL=30 ppm	Occupational Nasal histopathology (rat 13-wk inhalation tox.) Non-occupational Nasal histopathology (rat 13-wk inhalation tox.)	Occupational HEC=0.90 ppm UF=30 Non-occupational HEC=0.30 ppm UF=30 (100 for children)
Long-term (>6 months)			
USEPA (2008)	NOAEL=5 ppm LOAEL=20 ppm	Occupational Nasal histopathology (mouse chronic-onco.) Non-occupational Nasal histopathology (mouse chronic-onco.)	Occupational HEC=0.77 ppm UF=30 Non-occupational HEC=0.182 ppm UF=30
DPR-HHA (2015)	$BMCL_{10}$=6 ppm LOAEL=20 ppm	Occupational Nasal histopathology (mouse chronic-onco.) Non-occupational Nasal histopathology (mouse chronic-onco.)	Occupational HEC=0.59 ppm UF=30 Non-occupational HEC=0.20 ppm UF=30 (100 for children)

Oncogenicity			
USEPA (2008)	n/a	Occupational Bronchioloalveolar adenomas (mouse chronic-onco.)	Portal of entry, occup. AUR=$9.5x10^{-7}$ $(\mu g/m^3)^{-1}$
		Non-occupational Bronchioloalveolar adenomas (mouse chronic-onco.)	Portal of entry, non-occ. AUR=$4x10^{-6}$ $(\mu g/m^3)^{-1}$
DPR-HHA (2015)	n/a	Occupational Bronchioloalveolar adenomas (mouse chronic-onco.)	Portal of entry, occup. AUR=0.0059 $(ppm)^{-1}$ ($\approx 1.3x10^{-6}$ $(\mu g/m^3)^{-1}$) Systemic, occup. AUR=0.020 $(ppm)^{-1}$ ($\approx 4.4x10^{-6}$ $(\mu g/m^3)^{-1}$)
		Non-occupational Bronchioloalveolar adenomas (mouse chronic-onco.)	Portal of entry, non-occ. [a] AUR=0.018 $(ppm)^{-1}$ ($\approx 4x10^{-6}$ $(\mu g/m^3)^{-1}$) Systemic, non-occ. [a] AUR=0.062 $(ppm)^{-1}$ ($\approx 1.4x10^{-5}$ $(\mu g/m^3)^{-1}$)

Abbreviations: DPR-HHA, Human Health Assessment Branch, Dept. of Pesticide Regulation; HEC, human equivalent concentration; UF: uncertainty factor; dvp., developmental; AUR: air unit risk

[a] To calculate ambient cancer risks, the indicated non-occupational AURs were converted to cancer potency factors as specified above in section IV.B.3.d.. Those factors are 0.000014 $(\mu g/kg/day)^{-1}$ for portal of entry and 0.000048 $(\mu g/kg/day)^{-1}$ for systemic mode of action.

VI. REFERENCE CONCENTRATIONS (RFCS)

Reference concentrations (RfCs) are estimates of inhalation exposures to humans that are likely to be without appreciable risk of deleterious effects. Separate RfCs are generated for different age groups and exposure scenarios. These values are calculated by dividing the critical endpoint concentrations (expressed as human equivalent concentrations) by the uncertainty factors appropriate to the exposure scenarios evaluated. Uncertainty factors for 1,3-D were specified above in Table IV.4. Because the product of the uncertainty factors is equal to the target MOE, RfCs are simply the critical HEC divided by the target MOE.

Table VI.1 provides the RfC values for 1,3-D.

Table VI. 1 HECs, target MOEs and RfCs for the anticipated 1,3-D exposure scenarios

Exposure scenario	Age group	HEC [a] (ppm)	Target MOE [a] (ppm)	RfC (ppb)
Occupational scenarios				
Acute / short-term	Adult	33	30	1100
Seasonal	Adult	0.90	30	30
Annual	Adult	0.59	30	20
Non-occupational scenarios				
Acute / short-term	Adult	11	30	367
	Child	11	100	110
Seasonal	Adult	0.30	30	10
	Child	0.30	100	3
Annual	Adult	0.20	30	7
	Child	0.20	100	2

Abbreviations: HEC, human equivalent concentration; RfC, reference concentration

[a] HECs and target MOEs were calculated in section IV.A. above.

VII. CONCLUSIONS

Health risks to humans from inhalation exposure to 1,3-dichloropropene were assessed by combining toxicity studies conducted in laboratory animals with exposure projections for humans under non-occupational and occupational conditions. Since short-term, seasonal, annual and lifetime exposures were expected, corresponding risk values for each of these scenarios were calculated.

Non-oncogenic risk estimates. For non-oncogenic effects in adults, margins of exposure (MOEs) of 30 or greater were considered sufficient to protect human health. For non-oncogenic effects in children, MOEs of 100 or greater were considered sufficient to protect human health.

The MOE calculations were based on the following critical endpoint determinations in laboratory animals:

Acute / short-term $BMCL_{1\sigma}$: 49 ppm (body weight decrement in a rat 13-wk study)

Subchronic / seasonal $BMCL_{10}$: 16 ppm (nasal epithelial hyperplasia in a rat 13-wk study)

Chronic / annual $BMCL_{10}$: 6 ppm (nasal epithelia hyperplasia in a mouse 2-yr study)

MOEs for acute / short-term occupational scenarios were calculated by applying the critical occupational HEC of 33 ppm to the acute / short-term occupational exposure estimates. The highest risk acute / short-term risk occupation was tarp remover, which showed MOEs of 1 for various application types. Two additional occupational scenarios registered MOEs below the target of 30--- applicator (shallow shank without tarp; MOE = 17) and applicator (injection auger; MOE = 28). Occupational bystander (drip with tarp) showed an MOE of exactly 30.

Acute / short-term non-occupational MOEs were calculated by applying the critical HEC value of 11 ppm to the short-term exposure estimates. The lowest MOE of 20 was determined for a resident / bystander at the edge of a buffer zone for a shallow shank application. This value was below both the adult and child target MOEs of 30 and 100, respectively. No other MOEs were below 30. However, two of the remaining scenarios---near an application site at the edge of a buffer zone both for deep shank application and drip application---showed MOEs below 100 (MOE = 96 and 61, respectively). Two other scenarios---at the edge of the buffer zone for a tree and vine application and ambient---showed MOEs greater than 100.

MOEs for subchronic / seasonal occupational scenarios were calculated by applying the critical occupational HEC of 0.90 ppm to the seasonal occupational exposure estimates. The highest seasonal risk occupation was tarp remover, which showed MOEs of less than 1 for three different application types. Several additional occupational scenarios registered MOEs below the target MOE of 30, including applicator (shallow shank without tarp; MOE = 28), applicator (shallow shank with tarp; MOE = 9), applicator (deep shank without tarp; MOE = 13), applicator (deep

shank with tarp; MOE = 4), applicator (drip without tarp; MOE = 23), loader (shallow shank; MOE = 15), loader (deep shank; MOE = 7) and reentry worker (deep shank; MOE = 28).

MOEs for subchronic / seasonal risk for non-occupational scenarios were calculated by applying the critical HEC value of 0.30 ppm to the seasonal exposure estimates. The lowest MOE of 17 was determined for exposure at the edge of a buffer zone for a shallow shank application. This value was below both the adult and child target MOEs of 30 and 100, respectively. One additional seasonal exposure scenario showed a sub-30 MOE---near an application site, edge of buffer zone, deep shank (MOE = 22). Exposure near an application site, edge of buffer zone, drip gave an MOE of 60, indicating a seasonal health risk to children. This was also true for ambient exposure, with its seasonal MOE of 67.

MOEs for chronic / annual occupational scenarios were calculated by applying the critical occupational HEC of 0.59 ppm to the annual occupational exposure estimates. The highest annual occupational risk was tarp remover, which showed MOEs of less than 1 for three different application types. Several additional occupational scenarios registered MOEs below the target MOE of 30, including applicator (shallow shank with tarp; MOE = 18), applicator (deep shank without tarp; MOE = 14), applicator (deep shank with tarp; MOE = 4), loader (deep shank; MOE = 7) and reentry worker (deep shank; MOE = 25).

An MOE of 1000 was calculated using the critical non-occupational HED of 0.20 ppm for chronic / annual ambient risk. No other non-occupational exposure scenarios were anticipated.

Oncogenic risk estimates. For oncogenic effects, risk estimates less than the negligible risk standard of 10^{-6} were also considered sufficient to protect human health. These estimates were based on the appearance of bronchioloalveolar adenomas in males in a 2-year mouse inhalation study. Because the evidence did not overwhelmingly favor either a portal of entry or a systemic mode of oncogenic action, we opted to express cancer risk for both routes. Hence oncogenic risk ws calculated using the upper confidence limit (UCL) slope values---referred to as the air unit risk---of 0.0059 ppm^{-1} (portal of entry) and 0.020 ppm (systemic) for occupational scenarios, and 0.000014 [µg/kg/day]-1 (portal of entry) and 0.000048 [µg/kg/day]-1 for ambient lifetime exposure scenarios. These values were multiplied by the relevant lifetime exposures for the scenarios characterized in this analysis.

All of the occupational and ambient lifetime exposure scenarios showed oncogenic risk values that were above the negligible oncogenic risk standard of $1x10^{-6}$, regardless of assumed mode of action. Occupational cancer risk values for a portal of entry mode of action ranged between $7.1x10^{-6}$ (occupational bystander near an application site, 3 scenarios) and $1.7x10^{-2}$ (tarp remover, deep shank); for a systemic mode of action they ranged between $2.4x10^{-5}$ (occupational bystander near an application site, 3 scenarios) and $5.6x10^{-2}$ (tarp remover, deep shank) . Ambient cancer risks ranged between $2.30x10^{-6}$ (portal of entry, Mcable, 30-yr fixed, female) and $40.44x10^{-6}$ (systemic, HEE5CB, birth to age 70, low mobility).

Target MOEs and calculated MOEs for all exposure scenarios appear in Conclusion Table I. Oncogenic risk values for all occupational exposure scenarios appear in Conclusion Table II. Oncogenic risk values for ambient exposure scenarios appear in Conclusion Table III.

Conclusion Table I. Target MOEs and calculated MOEs for non-occupational and occupational 1,3-D exposure scenarios

Exposure scenario	Target MOE	Calculated MOE		
		Acute / short term	Seasonal	Annual
Occupational scenarios				
Applicator				
■ shallow shank w/o tarp	30 (adult)	122	28	61
■ shallow shank w/ tarp		39	9	18
■ deep shank w/o tarp		122	13	14
■ deep shank w/ tarp		39	4	4
■ drip w/o tarp		118	23	45
■ drip w/ tarp		143	50	98
■ injection auger		28	n/a	n/a
Loader				
■ shallow shank		47	15	31
■ deep shank		47	7	7
Tarp remover				
■ shallow shank		1	0.23	0.49
■ deep shank		1	0.11	0.11
■ drip		1	0.35	0.69
Reentry worker				
■ shallow shank		892	60	92
■ deep shank		892	28	25
■ drip		892	90	134
Occupational bystander				
■ shallow shank w/o tarp		17	750	952
■ deep shank w/o tarp		55	750	952
■ drip w/ tarp		30	750	952
Non-occupational scenarios				
Near application site, edge of buffer zone				
■ shallow shank	30 (adult); 100 (child)	20	17	n/a
■ deep shank		96	22	n/a
■ drip		61	60	n/a
■ tree & vine		120	n/a	n/a
Ambient		135	67	1000

Conclusion Table II. Oncogenic risk values for occupational 1,3-D exposure scenarios

Exposure scenario	Calculated oncogenic risk (negligible oncogenic risk standard = $1x10^{-6}$)	
Occupational scenarios		
	Portal of entry	**Systemic**
Applicator		
■ **shallow shank w/o tarp**	$3.2x10^{-5}$	$1.1x10^{-4}$
■ **shallow shank w/ tarp**	$1.0x10^{-4}$	$3.4x10^{-4}$
■ **deep shank w/o tarp**	$1.4x10^{-4}$	$4.6x10^{-4}$
■ **deep shank w/ tarp**	$4.3x10^{-4}$	$1.4x10^{-3}$
■ **drip w/o tarp**	$4.1x10^{-5}$	$1.4x10^{-4}$
■ **drip w/ tarp**	$1.9x10^{-5}$	$6.4x10^{-5}$
■ **injection auger**	n/a	n/a
Loader		
■ **shallow shank**	$5.9x10^{-4}$	$2.0x10^{-4}$
■ **deep shank**	$2.6x10^{-4}$	$8.8x10^{-4}$
Tarp remover		
■ **shallow shank**	$3.9x10^{-3}$	$1.3x10^{-2}$
■ **deep shank**	$1.7x10^{-2}$	$5.6x10^{-2}$
■ **drip**	$2.7x10^{-3}$	$9.2x10^{-3}$
Reentry worker		
■ **shallow shank**	$2.0x10^{-5}$	$6.8x10^{-5}$
■ **deep shank**	$7.1x10^{-5}$	$2.6x10^{-4}$
■ **drip**	$1.4x10^{-5}$	$4.8x10^{-5}$
Occupational bystander		
■ **shallow shank w/o tarp**	$1.9x10^{-6}$	$6.6x10^{-6}$
■ **deep shank w/o tarp**	$1.9x10^{-6}$	$6.6x10^{-6}$
■ **drip w/ tarp**	$1.9x10^{-6}$	$6.6x10^{-6}$

Conclusion Table III. Oncogenic risk values for ambient exposure scenarios

Ambient exposure scenarios – 95th percentile oncogenic risk				
	Portal of entry		Systemic	
	Male	Female	Male	Female
MCABLE				
With time away				
■ **Variable**	2.64×10^{-6}	2.49×10^{-6}	9.09×10^{-6}	8.56×10^{-6}
■ **30-yr fixed**	2.45×10^{-6}	2.30×10^{-6}	8.44×10^{-6}	7.91×10^{-6}
■ **50-yr fixed**	3.27×10^{-6}	3.04×10^{-6}	11.26×10^{-6}	10.44×10^{-6}
■ **70-yr fixed**	4.28×10^{-6}	3.97×10^{-6}	14.72×10^{-6}	13.64×10^{-6}
Without time away				
■ **Variable**	2.87×10^{-6}	2.69×10^{-6}	9.88×10^{-6}	9.23×10^{-6}
■ **30-yr fixed**	2.63×10^{-6}	2.49×10^{-6}	9.06×10^{-6}	8.57×10^{-6}
■ **50-yr fixed**	3.60×10^{-6}	3.31×10^{-6}	12.40×10^{-6}	11.39×10^{-6}
■ **70-yr fixed**	4.66×10^{-6}	4.27×10^{-6}	16.02×10^{-6}	14.68×10^{-6}
HEE5CB				
High mobility				
■ **Birth to age 30**	4.85×10^{-6}	4.75×10^{-6}	16.70×10^{-6}	16.34×10^{-6}
■ **Birth to age 50**	6.99×10^{-6}	6.76×10^{-6}	24.08×10^{-6}	23.25×10^{-6}
■ **Birth to age 70**	9.18×10^{-6}	8.77×10^{-6}	31.56×10^{-6}	30.17×10^{-6}
Intermediate mobility				
■ **Birth to age 30**	5.88×10^{-6}	6.97×10^{-6}	20.22×10^{-6}	23.99×10^{-6}
■ **Birth to age 50**	9.01×10^{-6}	8.16×10^{-6}	30.98×10^{-6}	28.08×10^{-6}
■ **Birth to age 70**	10.77×10^{-6}	10.6×10^{-6}	37.06×10^{-6}	35.45×10^{-6}
Low mobility				
■ **Birth to age 30**	6.25×10^{-6}	7.73×10^{-6}	21.51×10^{-6}	26.57×10^{-6}
■ **Birth to age 50**	10.43×10^{-6}	8.83×10^{-6}	35.89×10^{-6}	30.38×10^{-6}
■ **Birth to age 70**	11.75×10^{-6}	11.46×10^{-6}	40.44×10^{-6}	39.41×10^{-6}

REFERENCES

Andrews, C., and Patterson, G. 2000. Interim Guidance for Selecting Default Inhalation Rates for Children and Adults. Memorandum to Worker Health and Safety Branch Staff and Medical Toxicology Branch Staff, from Andrews, Chuck Chief, Worker Health and Safety Branch and Patterson, Gary, Chief, Medical Toxicology Branch, dated December 1. http://www.cdpr.ca.gov/docs/whs/memo/hsm00010.pdf.

ATSDR. 2008. Toxicological Profile for Dichloropropenes. *U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES, Public Health Service, Agency for Toxic Substances and Disease Registry*. 361. http://www.atsdr.cdc.gov/toxprofiles/tp40.pdf.

Banijamali, A. R., DeMatteo, V., and Sumner, S. J. 2003. A mechanism for the formation of bis-glutathione conjugates of propargyl alcohol. *Pest Manag Sci* 59:331-338.

Barry, T. 2015a. Review of Validation of SOFEA2 with 1,3-Dich1oropropene ambient monitoring data in Merced County California including mixing height modifications and border township effects. Regulatory Science and Government Affairs, Dow AgroSciences LLC, 9330 Zionsville Road, Indianapolis, Indiana 46268-1054. (DPR Vol. No. 50046-0230, Record No. 286007) 46.

Barry, T. 2014. Development of chloropicrin buffer zones - revised. http://www.cdpr.ca.gov/docs/whs/pdf/appendix_4_buffer_zones_memo.pdf.

Barry, T. A. 2015b. Personal communication to Miglena Wilbur, 2015.

Bartels, M. J., Hansen, S. C., Thornton, C. M., Brzak, K. A., Mendrala, A. L., Dietz, F. K., and Kastl, P. J. 2004. Pharmacokinetics and metabolism of ^{14}C-1,3-dichloropropene in the Fischer 344 rat and the $B_6C_3F_1$ mouse. *Xenobiotica* 34:193-213.

Basu, A. K., and Marnett, L. J. 1984. Molecular requirements for the mutagenicity of malondialdehyde and related acroleins. *Cancer Res* 44:2848-2854.

Beard, K. K., Murphy, P.G., Fontaine, D.D. and Weinberg, J.T. 1996. Monitoring of Potential Worker Exposure, Field Flux, and Off-Site Air Concentration During Chloropicrin Field Application. Lab Project Number: HEH 160. Unpublished study submitted by Chloropicrin Manufacturers Task Force. MRID 441492-01. .

Beauvais, S. 2006. Estimation of Exposure of Persons in California to Pesticide Products That Contain Carbofuran, pp. 68. Worker Health and Safety Branch, Division of Pest Management, Environmental Protection and Worker Safety, California Department of Food and Agriculture.

Beauvais, S. 2010 Chloropicrin Soil fumigation Occupational Exposure Data. Memorandum to, dated July 29, 2010. Sacramento, CA. http://www.cdpr.ca.gov/docs/whs/memo/hsm10005.pdf.

Beauvais, S. 2011. Respirator Assigned Protection Factors and Information on Compliance with Legal Requirements for Respirators. .

Beauvais, S. 2012. Estimation of Exposure of Persons in California to Pesticide Products That Contain Chloropicrin, pp. 149. Worker Health and Safety Branch, Division of Pest Management, Environmental Protection and Worker Safety, California Department of Food and Agriculture.

Beauvais, S. 2013. Review of Data Packages 261180 And 261979: Protocol for Residential Mobility Survey for Merced and Ventura Township Memorandum to Ann Hanger, Staff Environmental Scientist

Registration Branch, from Beauvais, Sheryl, Senior Toxicologist, Worker Health and Safety Branch, Worker Health and Safety Branch, dated December 9.

Bowden, D. H. 1983. Cell turnover in the lung. *Am Rev Respir Dis* 128:S46-48.

Breslin, W. J., Kirk, H. D., Streeter, C. M., Quast, J. F., and Szabo, J. R. 1987. Telone II Soil Fumigant: Two-generation inhalation reproduction study in Fischer 344 rats. (DPR Vol. No. 50046-030, Record No. 060676).

CDPR 1994. Interim Risk Assessment of 1,3-Dichlrorpropene for the Proposed Use of Telone (1994-1995), pp. 64. Health Assessment Section, Medical Toxicology Branch, Department of Pesticide Regulation, California Environmental Protection Agency.

CDPR 1997a. Risk Characterization Document of 1,3-Chloropropene, pp. 78.

CDPR 1997b. Risk Characterization Documetn of 1,3-Chloropropene, pp. 78.

CDPR 1999. Selection of the acute NOEL for Telone. Joyce Gee, dated October 8, 1999.

CDPR 2001. Suggested Permit Conditions for Using 1,3-Dichloropropene Pesticides (Fumigant). CDPR Enforcement Letter to, dated June 22, 2001.

CDPR 2002. Recommended Permit Conditions for Using 1,3-Dichloropropene Pesticides (Fumigant). CDPR Enforcement Letter to, dated September 12, 2002.

CDPR. 2002a. California management plan: 1,3-dichloropropene. *Department of Pesticide Regulation, California Environmental Protection Agency*. http://www.cdpr.ca.gov/docs/emon/pubs/envfate.htm.

CDPR. 2012. 1,3-Dichloropropene: Risk Characterization Document, Environmental Fate (author: Edgar Vidrio). *Environmental Monitoring Branch, Department of Pesticide Regulation, California Environmental Protection Agency.* http://www.cdpr.ca.gov/docs/emon/pubs/fatememo/13-d.pdf.

CDPR. 2014. Subsection C.7.1 1,3-Dichloropropene Pesticides (Field Fumigant) Recommended Permit Conditions. In *Pesticide Use Enforcement Program Standards Compendium Volume 3: Restricted Materials and Permitting*: State of California, Department of Pesticide Regulation, 1001 I Street, P.O. Box 4015, Sacramento, California 95812-4015, pp. 12.

CDPR. 2015a. Pesticide Illness Query, 1992-2011. Query of California Pesticide Illness Query (CalPIQ) database, 5-28-2015 by J. McKenzie, Human Health Assessment Branch. URL.

CDPR. 2015b. Pesticide Illness Surveillance Program (PISP) Database User Documentation/Dictionary. (May 18, 2015). 71.

CDPR 2015c. Pesticide Use Enforcement Program Standards Compendium Volume 3: Restricted Materials and Permitting, pp. 458. State of California, Department of Pesticide Regulation, 1001 I Street, P.O. Box 4015, Sacramento, California 95812-4015.

CDPR. 2015d. Pesticide Use Report-1,3-Dichloropropene 1998-2007. Query of Pesticide Use Report database, 4-18-2015 by J. McKenzie, Human Health Assessment Branch. URL.

CDPR. 2015e. Pesticide Use Report-1,3-Dichloropropene 2008-2012. Query of Pesticide Use Report database, 4-18-2015 by J. McKenzie, Human Health Assessment Branch. URL.

Clary, T., and Ritz, B. 2003. Pancreatic cancer mortality and organochlorine pesticide exposure in California, 1989-1996. *American journal of industrial medicine* 43:306-313.

Coate, W. B. 1979. 90-day inhalation toxicity study in rats and mice: Telone II. Hazleton Laboratories America for Dow Chemical Co. (DPR Vol. No. DPR Vol. #50046-010, Record No. 36551).

Cohen, S. M., Garland, E. M., St John, M., Okamura, T., and Smith, R. A. 1992. Acrolein initiates rat urinary bladder carcinogenesis. *Cancer Res* 52:3577-3581.

Connors, T. F., Stuart, J. D., and Cope, J. B. 1990. Chromatographic and mutagenic analyses of 1,2-dichloropropane and 1,3-dichloropropylene and their degradation products. *Bull Environ Contam Toxicol* 44:288-293.

Cox, C. 1992. 1,3-Dichloropropene. *J. Pesticide Reform* 12.

Cracknell, S., Jackson, G., and Hardy, C. 1987. Telone* II (1,3-dichloropropene) acute inhalation study in rats, 4 hour exposure. . March 25, 1987. Report designation DWC/484. No DPR Volume or record number. . (DPR Vol.

Creedy, C. L., Brooks, T. M., Dean, B. J., Hutson, D. H., and Wright, A. S. 1984. The protective action of glutathione on the microbial mutagenicity of the Z- and E-isomers of 1,3-dichloropropene. *Chem Biol Interact* 50:39-48.

Cryer, S., Wesenbeeck, I. v., and Reiss, R. 2015. Technical Details for SOFEA.

Cryer, S. A., and van Wesenbeeck, I. 2000a. Termporal And Spatial Distributions of Township Air Concentrations of 1,3-Dichloropropene Resulting from Treated Fields In The California Tree and Vine Market. Dow Agrosciences Indianapolis, In (5796) (DPR Vol. No. 50046-0151, Record No. 177308) 110.

Cryer, S. A., and Van Wesenbeeck, I. J. 2000b. Predicted Township Wide Air Concentrations Of 1,3-Dichloropropene Resulting From Tree And Vine Applications In California. Dow Agrosciences Global Environmental Chemistry Laboratory-Indianapolis, IN (5796) Dow AgroSciences (DPR Vol. No. 50046-0148 Record No. 174339) 98.

Cryer, S. A., van Wesenbeeck, I. J., and Houtma, B. A. 2004. Predicting Soil Fumigant Acute, Sub-chronic, and Chronic Air Concentrations Under Diverse Agronomic Practices.

Damsch, S., Eichenbaum, G., Looszova, A., Lammens, L., Feyen, B., Van den Bulck, K., Knight, E., Kelley, M., and Tonelli, A. 2011a. Unexpected nasal changes in rats related to reflux after gavage dosing. *Tocologic Pathology* 39:337-347.

Damsch, S., Eichenbaum, G., Tonelli, A., Lammens, L., Van den Bulck, K., Feyen, B., Vandenberghe, J., Megens, A., Knight, E., and Kelley, M. 2011b. Gavage-related reflux in rats: identification, pathogenesis, and toxicological implications (review). *Tocologic Patholog* 39:348-360.

DAS 2011. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene in Calfornia from 1995 to 2010 (CD). IN (5796) Dow AgroSciences (DPR Vol. No. 50046-0196). .

DAS 2012. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene in Calfornia from 1995 to 2011 (CD). IN (5796) Dow AgroSciences (DPR Vol. No. 50046-0199). .

DAS 2013. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene (1,3-D) in Calfornia, 1995-2012 (Township Cap Data) (CD) IN (5796) Dow AgroSciences (DPR Vol. No. 50046-0205). .

DAS 2014. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene (1,3-D) in California, 1995-2012 (Township Cap Data) (CD) IN (5796) Dow AgroSciences (DPR Vol. No. 50046-0211). . In *Dow Agrosciences LLC* Dow Agrosciences Indianapolis, In (5796)

DAS 2015a. 1,3-Dichloropropene Historical Use Data in California from 1995-2014 (CD). Dow Agrosciences LLC Dow Agrosciences Indianapolis, In (5796) 50046-0224.

DAS 2015b. Power-Point Presentation: Exposure and Risk Assessment for 1,3-D in California: Background, Refined Bystander Exposure & Risk Analysis: (1) Air Dispersion Modeling (SOFEA), (2) Residency-Mobility, and (3) Probabilistic Exposure Modeling (MCABLE). Dow AgroSciences.

DeMaster, E. G., Dahlseid, T., and Redfern, B. 1994. Comparative oxidation of 2-propyn-1-ol with other low molecular weight unsaturated and saturated primary alcohols by bovine liver catalase in vitro. *Chem Res Toxicol* 7:414-419.

Dix, K. J., Coleman, D. P., Fossett, J. E., Gaudette, N. F., Jr., Stanley, A. P., Thomas, B. F., and Jeffcoat, A. R. 2001. Disposition of propargyl alcohol in rat and mouse after intravenous, oral, dermal and inhalation exposure. *Xenobiotica* 31:357-375.

Dorman, D. C., Struve, M. F., Wong, B. A., Marshall, M. W., and Gross, E. A. 2008. Repiratory tract responses in male rats following subchronic acrolein inhalation. *Inhalation Toxicology* 20:205-216.

Dow Agrosciences. 2011. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene in California from 1995 to 2010 (CD). (DPR Vol. No. 50046-0196, Record No. 258557).

Dow Agrosciences. 2012. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene in California from 1995 to 2011 (CD). Dow Agrosciences Indianapolis, In (5796) Dow Agrosciences LLC (DPR Vol. No. 50046-0199, Record No. 265298).

Dow Agrosciences. 2014. Historical Use Data of Adjusted Pounds of 1,3-Dichloropropene in California from 1995 to 2013 (CD). Dow Agrosciences Indianapolis, In (5796) Dow Agrosciences LLC (DPR Vol. No. 50046-0211, Record No. 277412).

Dow Agrosciences. 2015. 1,3-Dichloropropene Historical Use Data in California from 1995-2014 (CD). Dow Agrosciences Indianapolis, In (5796) Dow Agrosciences LLC (DPR Vol. No. 50046-0224, Record No. 283299).

Dow Chemical Company. 1985a. Evaluation of Telone II in the Rat Hepatocyte Unscheduled DNA Synthesis Assay. . (DPR Vol. No.???, Record No.???).

Dow Chemical Company. 1985b. Evaluation of Telone II Soil Fumigant in the Mouse Bone Marrow Micronucleus Test. (DPR Vol. No. 50046-010, Record No. 036560).

Dow Chemical Company. 1986. The Evaluation of Telone II Soil Fumigant in the CHO Cell/HGPRT Forward Mutation Assay. (DPR Vol. No. 50046-019, Record No. 042045).

Driver, J. H., Ross, J. H., Cochran, R. C., Holden, L., VanWesenbeeck, I., and Price, P. S. 2014a. Evaluation Of Potential Human Health Effects Associated With The Agricultural Uses Of 1,3-D: Refined Stochastic Risk Analysis. Dow AgroSciences LLC, 9330 Zionsville Road, Indianapolis, Indiana 46268, USA. (DPR Vol. No. 50046-0220, Record No. 282371) 120.

Driver, J. H., Ross, J. H., Cochran, R. C., Holden, L., VanWesenbeeck, I., and Price, P. S. 2015. Evaluation Of Potential Human Health Effects Associated With The Agricultural Uses of 1,3-D: Refined Stochastic Risk Analysis Amended Report. risksciences.net, LLC, Manassas, VA 20111. (DPR Vol. No. 50046-0231, Record No. 286008) 120.

Driver, J. H., Wesenbeeck, I. v., and Price, P. S. 2014b. 1,3-D Probabilistic Risk Analysis. Dow AgroSciences.

Eder, E., Espinosa-Gonzalez, J., Mayer, A., Reichenberger, K., and Boerth, D. 2006. Autoxidative activation of the nematocide 1,3-dichloropropene to highly genotoxic and mutagenic derivatives: consideration of genotoxic/carcinogenic mechanisms. *Chem Res Toxicol* 19:952-959.

Eder, E., Henschler, D., and Neudecker, T. 1982b. Mutagenic properties of allylic and alpha, beta-unsaturated compounds: consideration of alkylating mechanisms. *Xenobiotica* 12:831-848.

Eder, E., Hoffman, C., Bastian, H., Deininger, C., and Scheckenbach, S. 1990. Molecular mechanisms of DNA damage initiated by alpha, beta-unsaturated carbonyl compounds as criteria for genotoxicity and mutagenicity. *Environ Health Perspect* 88:99-106.

Eder, E., Neudecker, T., Lutz, D., and Henschler, D. 1980. Mutagenic potential of allyl and allylic compounds. Structure-activity relationship as determined by alkylating and direct in vitro mutagenic properties. *Biochem Pharmacol* 29:993-998.

Eder, E., Neudecker, T., Lutz, D., and Henschler, D. 1982a. Correlation of alkylating and mutagenic activities of allyl and allylic compounds: standard alkylation test vs. kinetic investigation. *Chem Biol Interact* 38:303-315.

EFSA. 2009. Conclusion on the peer review of the pesticide risk assessment of the active substance (EZ)-1,3-dichloropropene. 7, no. 10. 1-102.

Epstein, J., Rosenthal, R. W., and Ess, R. J. 1955. Use of γ-(4-nitrobenzyl)pyridine as analytical reagent for ethylenimines and alkylating agents. *Analytical Chemistry* 27:1435-1439.

Farber, E. 1973. Carcinogenesis - cellular Evolution as a unifying thread: Presidential address. *Cancer Research* 33:2537-2550.

Fedtke, N., Certa, H., Ebert, R., and Wiegand, H. J. 1994. Species differences in the biotransformation of ethyl chloride. I. Cytochrome P450-dependent metabolism. *Arch Toxicol* 68:158-166.

Frank, J. P. 2009. Review of Proposed Update to the California Management Plan. http://apps.cdpr.ca.gov/whsrpts/hsmemo/hsmem_adv_action.cfm

http://www.cdpr.ca.gov/docs/whs/memo/hsm09002.pdf.

Fukushima, S., Kinoshita, A., Puatanachokchai, R., Kushida, M., Wanibuchi, H., and Morimura, K. 2005. Hormesis and dose-response-mediated mechanism in carcinogenesis: evidence for a threshold in carcinogenicity of non-genotoxic carcinogens. *Carcinogenesis* 26:1835-1845.

Galloway, S. M., Armstrong, M. J., Reuben, C., Colman, S., Brown, B., Cannon, C., Bloom, A. D., Nakamura, F., Ahmed, M., Duk, S., and et al. 1987. Chromosome aberrations and sister chromatid exchanges in Chinese hamster ovary cells: evaluations of 108 chemicals. *Environmental and molecular mutagenesis* 10 Suppl 10:1-175.

Ghia, M., Robbiano, L., Allavena, A., Martelli, A., and Brambilla, G. 1993. Genotoxic activity of 1,3-dichloropropene in a battery of in vivo short-term tests. *Toxicology and applied pharmacology* 120:120-125.

Gillis, M. J., and Dowling, K. C. 1998. Effect of broadcast and row application methods on 1,3-dichloropropene emissions. Indianapolis, Indiana 46268: Dow AgroSciences HEA95177. (DPR Vol. No. 50046-127, Record No. 167938).

Gollapudi, B. B., and Cieszlak, F. S. 1997. Telone II Soil Fumigant: Evaluation in an in vivo assay for gene mutagens using transgenic Big Blue Mice. (DPR Vol. No. 50046-120, Record No. 162475).

Gollapudi, B. B., Cieszlak, F. S., Day, S. J., and Carney, E. W. 1998. Dominant lethal test with rats exposed to 1,3-dichloropropene by inhalation. *Environmental and molecular mutagenesis* 32:351-359.

Gollapudi, B. B., Cieszlak, F. S., and Lick, S. J. 1997. Telone* II soil fumigant (cis/trans 1,3-dichloropropene): inhalation dominant lethal mutagenicity study in the CD (Sprague-Dawley derived) rat. (DPR Vol. No. 50046-0115, Record No. 162466).

Gosselin, P. 2001a. Interim Changes to Buffer Zones for 1,3Dichloropropene (Telone) Applications to Permanent Crops (Tree and Vine). Memorandum to Paul Helliker, from Paul Gosselin, Acting Chief Deputy Director, Department of Pesticide Regulation, dated January 5. Sacramento, CA.

Gosselin, P. 2001b. Managing 1,3-Dichloropropene (Telone) Chronic Risks. Memorandum to Tobi L. Jones, PhD, Ron Oshima, and Douglas Y. Okumura, Department of Pesticide Regulation, from Paul Gosselin, Acting Chief Deputy Director, Department of Pesticide Regulation, dated April 9. Sacramento, CA.

Gosselin, P. 2002. Risk Management Steps for 1,3-Dichloropropene (1,3D). . Memorandum to Tobi L. Jones, PhD, Ron Oshima, and Douglas Y. Okumura, Department of Pesticide Regulation, from Paul Gosselin, Acting Chief Deputy Director, Department of Pesticide Regulation, dated March 26. Sacramento, CA.

Haies, D. M., Gil, J., and Weibel, E. R. 1981. Morphometric study of rat lung cells. I. Numerical and dimensional characteristics of parenchymal cell population. *Am Rev Respir Dis* 123:533-541.

Haworth, S., Lawlor, T., Mortelmans, K., Speck, W., and Zeiger, E. 1983. Salmonella mutagenicity test results for 250 chemicals. *Environmental Mutagenesis* 5:3-49.

Heddle, J. A., Dean, S., Nohmi, T., Boerrigter, M., Casciano, D., Douglas, G. R., Glickman, B. W., Gorelick, N. J., Mirsalis, J. C., Martus, H. J., Skopek, T. R., Thybaud, V., Tindall, K. R., and Yajima, N. 2000. In vivo transgenic mutation assays. *Environmental and molecular mutagenesis* 35:253-259.

Herrett, R. A., and Kurtz, A. N. 1963. cis-3-Chloroacrylic Acid: A New Cotton Defoliant and Crop Desiccant. *Science* 141:1192-1193.

Houtman, B. A. 1992. Measurement of Off-Site Air Concentrations of 1,3-Dichloropropene Following Application of Telone Soil Fumigant - Phase I (Interim Report). DowElanco & Co. Indianapolis, In (4757) DowElanco & Co. Indianapolis, In (4757) (DPR Vol. No. 50046-0070, Record No. 121823) 68.

Houtman, B. A. 1993. Evaluation Of 1,3-Dichloropropene Worker Exposure Aassociated With Telone Soil Fumigant Loading, Application And Re-Entry (Final Report, Includes Summary) DowElanco North American Environmental Chemistry Laboratory Indianapolis, In (5324) DowElanco (DPR Vol. No. 50046-0071, Record No. 126461) 167.

Hoyes, K. P., Wadeson, P. J., Sharma, H. L., Hendry, J. H., and Morris, I. D. 1998. Mutation studies in lacI transgenic mice after exposure to radiation or cyclophosphamide. *Mutagenesis* 13:607-612.

Jeffrey, M. M. 1987. Telone II soil fumigant: dermal sensitization potential in the Hartley albino guinea pig. Midland, MI: Dow Chemical Co., Laboratory study #HET M-0039930017E. (DPR Vol. No. 50046-0032, Record No. 062073).

John, J. A., Kloes, P. M., Calhoun, L. L., and Young, J. T. 1983. Telone II: Inhalation teratology study in Fischer 344 rats and New Zealand White rabbits. (DPR Vol. No. 50046-0010, Record No. 36561 (the rat part of the study)).

Johnson, B. 2007. Simulation of Concentrations and Exposure Associated with Dow Agrosciences-Proposed Township Caps for Ventura County for 1,3-Dichloropropene. Momerandum to, dated March 27. http://www.cdpr.ca.gov/docs/emon/pubs/ehapreps/analysis_memos/ventur_telone.pdf.

Johnson, B. 2009a. Calculation Of Screening Concentrations for 1,3-Dichloropropene. Memorandum to Frank, Joseph P. , Worker Health and Safety Branch, from Johnson, Bruce Research Scientist III, Environmental Monitoring Branch, dated December 2. http://ext-testsite/docs/emon/pubs/ehapreps/analysis_memos/4467_johnson.pdf.

Johnson, B. 2009b. Subchronic 1,3-Dichloropropene Air Concentration Estimates. Memorandum to Frank, Joseph P. , Worker Health and Safety Branch, from Johnson, Bruce, Environmental Monitoring Branch, dated December 17. http://www.cdpr.ca.gov/docs/emon/pubs/analysmemos.htm.

Johnson, B. 2014a. Comparison of One-Year Township Monitoring Results From Merced To SOFA Simulation Results. Memorandum to Segawa, Randy Environmental Monitoring Branch, from Johnson, Bruce Research Scientist III, Environmental Monitoring Branch, dated August 20. http://www.cdpr.ca.gov/docs/emon/pubs/analysmemos.htm.

Johnson, B. 2014b. Evaluation: Validation of SOFEA2 with 1,3-Dichloropropene ambient monitoring data in Merced County California – REVISED report, pp. 31.

Johnson, B., and Powell, S. 2005. Interim Statewide Caps Analysis For 1,3-Dichloropropene. Memorandum to, dated December 28. http://www.cdpr.ca.gov/docs/whs/memo/hsm05014.pdf.

Johnson, B., Powell, S., and Reed, N.-m 2001. DAS Request to Shorten Buffer Zone Duration for Tree and Vine Applications from Seven to Four Days. Memorandum to John S. Sanders, Environmental Monitoring Branch, Department of Pesticide Regulation, from Bruce Johnson, Sally Powell, Nu-May Ruby Reed, Senior Environmental Research Scientist, Senior Environmental Research Scientist, and Staff Toxicologist, Department of Pesticide Regulation, dated March 27.

Johnson, B., and Spurlock, F. C. 2012. A method for estimating near-field air concentrations following tarp cutting for broadcast applications. Environmental Monitoring Branch, Department of Pesticide Regulation, California Environmental Protection Agency. 1001 I street. P.O. Box 4015, Sacramento, CA 95812-4015. March 9, 2012. http://www.cdpr.ca.gov/docs/emon/pubs/ehapreps/analysis_memos/2369-segawa.pdf.

Johnson, B. a. K., W. 2002. Flux Summary Database. Environmental Monitoring Branch, Department of Pesticide Regulation, California Environmental Protection Agency, 1001 I St., P.O. Box 4015, Sacramento, CA 95812-4015.

Kaplan, W. 2014. Residential Mobility Survey for Merced and Ventura Townships. California Survey Research Services, Inc., Van Nuys, CA: Dow AgroSciences LLC, 9330 Zionsville Road, Indianapolis, Indiana 46268, USA. (DPR Vol. No. 50046-0215 Record No. 281163) 102.

Kevekordes, S., Gebel, T., Pav, K., Edenharder, R., and Dunkelberg, H. 1996. Genotoxicity of selected pesticides in the mouse bone-marrow micronucleus test and in the sister-chromatid exchange test with human lymphocytes in vitro. *Toxicol Lett* 89:35-42.

Kim, S. J., Jung, Y. S., Yoon, M. Y., and Kim, Y. C. 2007. Comparative effects of dimethylsulfoxide on metabolism and toxicity of carbon tetrachloride and dichloromethane. *J Appl Toxicol* 27:25-31.

Kitchin, K. T., and Brown, J. L. 1989. Biochemical effects of three carcinogenic chlorinated methanes in rat liver. *Teratog Carcinog Mutagen* 9:61-69.

Kitchin, K. T., and Brown, J. L. 1994. Dose-response relationship for rat liver DNA damage caused by 49 rodent carcinogens. *Toxicology* 88:31-49.

Kitchin, K. T., Brown, J. L., and Kulkarni, A. P. 1991. Ornithine decarboxylase induction and DNA damage as a predictive assay for potential carcinogenicity. *Prog Clin Biol Res* 369:137-144.

Kitchin, K. T., Brown, J. L., and Kulkarni, A. P. 1992. Predictive assay for rodent carcinogenicity using in vivo biochemical parameters: operational characteristics and complementarity. *Mutat Res* 266:253-272.

Kitchin, K. T., Brown, J. L., and Kulkarni, A. P. 1993. Predicting rodent carcinogenicity of halogenated hydrocarbons by in vivo biochemical parameters. *Teratog Carcinog Mutagen* 13:167-184.

Klaunig, J. E., Gehen, S. C., Wang, Z., Klein, P. J., and Billington, R. 2015. Mechanism of 1,3-dichloropropene-induced rat liver carcinogenesis. *Toxicological sciences : an official journal of the Society of Toxicology* 143:6-15.

Kloes, P. M., Chalhoun, L. L., Young, J. T., and John, J. A. 1983. Telone II: inhalation teratology probe study in Fischer 344 rats and New Zealand White rabbits. (DPR Vol. No. 50046-0032, Record No. 06073).

Knuteson, J. A., Dixon-White, H. E., and Petty, D. G. 1995. Field volatility of 1,3-dichloropropene in San Joaquin Valley California. Indianapolis, Indiana 46808: DowElanco ENV93063. (DPR Vol. No. 50046-088, Record No. 134784).

Knuteson, J. A., and Dolder, S. C. 2000. Feild volatility of 1,3-dichloropropene and chloropicrin from shallow drip irrigtaion application of TELONE C-35 (InLine) to strawberry beds covered with VIF Tarp. Indianapolis, Indiana 46268-1054: Dow AgroSciences LLC 980070.01. (DPR Vol. No. 50046-153, Record No. 178247).

Knuteson, J. A., Petty, D. G., and Shurdut, B. A. 1992a. Field volatility of 1,3-dichloropropene in Salinas Valley California. Midland, MI 48674: DowElanco ENV91011. (DPR Vol. No. 50046-0067, Record No. 120011).

Knuteson, J. A., Petty, D. G., and Shurdut, B. A. 1992b. Field Volatility of 1,3-dichloropropene in the Imperial Valley of southern California. Midland, MI 48641-1706: DowElanco ENV91001. (DPR Vol. No. 50046-0053, Record No. 113745).

Konishi, Y., Kawabata, A., Denda, A., Ikeda, T., Katada, H., and Maruyama, H. 1980. Forestomach tumors induced by orally administered epichlorohydrin in male Wistar rats. *Gann* 71:922-923.

Lawlor, T. E. 1996. Evaluation of 1,3-dichloropropene for mutagenic potential in Salmonella in the presence of mouse lung homogenate (S9). In *Corning Hazleton Inc. (CHV), CHV No 17037.04-404*.

Leahy, B. 2014. Suspension of Exceptions to Township Limitations on Use of 1,3-Dichloropropene (1,3-D). Memorandum to Verder-Carlos, Marylou N. , Dpartment of Pesticide Regulation, from Leahy, Brian, Director, D~artment of Pesticide Regulation, dated February 19. http://www.cdpr.ca.gov/docs/emon/methbrom/telone/susp_of_excptn_telone.pdf.

Levin, D. E., Hollstein, M., Christman, M. F., Schwiers, E. A., and Ames, B. N. 1982b. A new *Salmonella* tester strain (TA102) with A-T base pairs at the site of mutation detects oxidative mutagens. *Proc Natl Acad Sci U S A* 79:7445-7449.

Levin, D. E., Yamasaki, E., and Ames, B. N. 1982a. A new Salmonella tester strain, TA97, for the detection of frameshift mutagens. A run of cytosines as a mutational hot-spot. *Mutat Res* 94:315-330.

Lewis, C. M. 2012. Chloropicrin Risk Characterization Document, pp. 118, Sacramento, CA.

Liu, J. 2014. Dichloropropene. In *Encyclopedia of Toxicology*, edited by P. Wexler: Academic Press, pp. 102-103.

Lomax, L. G., Calhoun, L. L., Stott, W. T., and Frauson, L. E. 1987. Telone II soil fumigant: 2-year inhalation chronic toxicity-oncogenicity study in rats. (DPR Vol. No. 50046-0031, Record No. 060677).

Loveday, K. S., Lugo, M. H., Resnick, M. A., Anderson, B. E., and Zeiger, E. 1989. Chromosome aberration and sister chromatid exchange tests in chinese hamster ovary cells in vitro: II. Results with 20 chemicals. *Environmental and molecular mutagenesis* 13:60-94.

Markovitz, A., and Crosby, W. H. 1984. Chemical carcinogenesis: a soil fumigant, 1,3-dichloropropene, as possible cause of hematologic malignancies. *Archives of Internal Medicine* 144:1409-1411.

McCall, P. J. 1987. Hydrolysis of 1,3-dichloropropene in dilute aqueous solution. *Pesticide Science* 19:235-242.

McCall, S. N., Jurgens, P., and Ivanetich, K. M. 1983. Hepatic microsomal metabolism of the dichloroethanes. *Biochem Pharmacol* 32:207-213.

McFee, A. F. 1989. Genotoxic potency of three quinoline compounds evaluated in vivo in mouse marrow cells. *Environmental and molecular mutagenesis* 13:325-331.

Mhatre, S. S., Chetty, K. G., and Pradhan, D. S. 1983. Uncoupling of oxidative phosphorylation in rat liver mitochondria following the administration of dimethyl sulphoxide. *Biochem Biophys Res Commun* 110:325-331.

Moore, M. M., and Doerr, C. L. 1990. Comparison of chromosome aberration frequency and small-colony TK-deficient mutant frequency in L5178Y/TK(+/-)-3.7.2C mouse lymphoma cells. *Mutagenesis* 5:609-614.

Morgan, E. T., Koop, D. R., and Coon, M. J. 1982. Catalytic activity of cytochrome P-450 isozyme 3a isolated from liver microsomes of ethanol-treated rabbits. Oxidation of alcohols. *J Biol Chem* 257:13951-13957.

Myhr, B. C., Caspary, W. J., and Holden, H. E. 1991. Chemical mutagenesis at the thymidine kinase locus in L5178Y mouse lymphoma cells: Results for 31 coded compounds in the national toxicology program. *Environmental and molecular mutagenesis* 18:51-83.

Neudecker, T., and Henschler, D. 1986. Mutagenicity of chloroolefins in the Salmonella/mammalian microsome test. III. Metabolic activation of the allylic chloropropenes allyl chloride, 1,3-dichloropropene, 2,3-dichloro-1-propene, 1,2,3-trichloropropene, 1,1,2,3-tetrachloro-2-propene and hexachloropropene by S9 mix via two different metabolic pathways. *Mutat Res* 170:1-9.

Neuman, H.-G. 2009. Risk assessment of chemical carcinogens and thresholds. *Critical Reviews in Toxicology* 39:449-461.

Nicolini, C., Robbiano, L., Pino, A., Maura, A., Finollo, R., and Brambilla, G. 1985. Higher sensitivity for the detection of chemically-induced DNA damage: role of DNA unfolding in determining alkaline elution rate. *Carcinogenesis* 6:385-389.

Nitschke, K. D., Crissman, J. W., and Schuetz, D. J. 1990a. Ci5-1,3-Dichloropropene: Acute Inhalation Study With Fischer 344 Rats. The Toxicology Research Laboratory, Health and Environmental Sciences, The Dow Chemical Company, Midland, Michigan 48674. (DPR Vol.

Nitschke, K. D., Crissman, J. W., and Schuetz, D. J. 1990b. Cis-1,3-dichloropropene: acute inhalation study with Fischer 344 rats. (DPR Vol. No. 50046-217, Record No. 282091).

Nomura Sogo Research Institute. 1978. Mutagenicity of 1,3-Dichloropropene in Bacteria Test System. (DPR Vol. No. 50046-010, Record No. 036556).

NTP. 1985. Toxicology and carcinogenesis studies of Telone II in F344/N rats and B6C3F1 mice NTP Technical Report Series #269. (May 1985).

OEHHA 2012. Notice of Adoption of Air Toxics Hot Spots Program Risk Assessment Guidelines: Revised Technical Support Document for Exposure Assessment And Stochastic Analysis In *Air Toxics Hot Spots Program*.

OSHA 2009. Assigned Protection Factors for the Revised Respiratory Protection Standard.

Park, S. B., Osterloh, J. D., Vamvakas, S., Hashmi, M., Anders, M. W., and Cashman, J. R. 1992. Flavin-containing monooxygenase-dependent stereoselective S-oxygenation and cytotoxicity of cysteine S-conjugates and mercapturates. *Chem Res Toxicol* 5:193-201.

Patel, B. V., Wilson, M. R., and Takata, M. 2012. Resolution of acute lung injury and inflammtion - a translational mouse model. *European Respiratory Journal* 39:1162-1170.

Patel, J. M., Wood, J. C., and Leibman, K. C. 1980. The biotransformation of allyl alcohol and acrolein in rat liver and lung preparations. *Drug Metab Dispos* 8:305-308.

Poet, T. S., Kirman, C. R., Bader, M., Thriel, C. v., Gargas, M. L., and Hinderliter, P. M. 2010. Quantitative risk analysis for N-methyl pyrrolidone using physiologically based pharmacokinetic and benchmark dose modeling. *Toxicological Sciences* 113:468-482.

Powell, S. 2000. Acute Exposure Values For Tree And Vine Applications Of 1,3-Dichloropropene With Reduced Buffer Zone. Memorandum to, dated November 13. http://www.cdpr.ca.gov/docs/whs/memo/hsm00016.pdf.

Powell, S. 2006. Estimating Residential Mobility For The Assessment Of Lifetime Exposure To 1,3-Dichloropropene. Frank, Joseph P. , Worker Health and Safety Branch, from Senior Environmental Research Scientist, Worker Health and Safety Branch, dated December 31. http://www.cdpr.ca.gov/docs/whs/memo/hsm06015.pdf.

Purchase, I. F. H., and Auton, T. R. 1995. Thresholds in chemical carcinogenesis. *Regulatory Toxicology and Pharmacology* 22:199-205.

Ray, S. I., and Houtman, B. A. 1995. Simulation Of 1,3-Dichloropropene Air Concentrations In Fresno County, California Following Tree And Vine Applications Of Telone II (Includes Protocol). Modeling and Information Sciences Laboratory, DowElanco 9330 Zionsville Road, Indianapolis, Indiana 46268-1053: DowElanco North American Environmental Chemistry Laboratory Indianapolis, In (5324) (DPR Vol. No. 50046-0143, Record No. 171265) 40.

Rick, D. L., and McCarty, L. P. 1988. The determination of the odor threshold of Telone II soil fumigant by a new method. *Applied Industrial Hygiene* 3:299-302.

Rotondaro, A., and Van Wesenbeeck, I. 2012a. Monitoring of Cis- And Trans-1,3-Dichloropropene In Air in 9 High 1,3-Dichloroprepene Use Townships Merced County, California (2 Pts.). Dow Agrosciences Indianapolis, IN (5796) Dow Agrosciences (DPR Vol. No. 50046-0200, Record No. 266848) **Note: Same as 50046-0203, #269511 (MW, 01/26/15).** 405.

Rotondaro, A., and van Wesenbeeck, I. 2012b. Monitoring of Cis- and Trans-1,3-Dichloropropene in Air In 9 High 1,3-Dichloropropene Use Townships Merced County, California. Multiple Sources: Combined Reports From Various Laboratories (4935) Dow Agrosciences LLC (DPR Vol. No. 50046-0203, Record No. 269511) 405.

Rotondaro, A. U. s. s. b. t. C. M. T. F., Mojave, California. Laboratory Study ID PRS02004. MRID 464202-01. . 2004. Monitoring of Chloropicrin Emissions from Field and Greenhouse Drip Irrigation Applications, and Implied Worker Inhalation Exposure from Applications of Chloropicrin by Shank Injection, Drip Irrigation Systems and at Tree Replant Sites. . DPR Data Volume 199-112, no.

Sanborn, J. R., and Powell, S. 1994. Human Exposure Assessment for 1,3-dichloropropene, pp. 29. California Environmental Protection Agency, Department of Pesticide Regulation, Worker Health and Safety Branch, 1020 N Street, Sacramento California 95814.

Sarkar, B. 2010. A classification of tarps by cluster analysis based on mass transfer coefficient data contained in analytical chemistry branch laboratory report entitled "*Agricultural Tarp Permeability to Fumigants.*". (August 27, 2010). DP Barcode: 381403.

Schneider, M., Quistad, G. B., and Casida, J. E. 1998a. 1,3-Dichloropropene epoxides: intermediates in bioactivation of the promutagen 1,3-dichloropropene. *Chem. Res. Toxicol.* 11:1137-1144.

Schneider, M., Quistad, G. B., and Casida, J. E. 1998b. N^2,7-bis(1-hydroxy-2-oxopropyl)-2'-deoxyguanosine: identical noncyclic adducts with 1,3-dichloropropene epoxides and methylglyoxal. *Chem Res Toxicol* 11:1536-1542.

Sells, D. M., Brix, A. E., Nyska, A., Jokinen, M. P., Orzech, D. P., and Walker, N. J. 2007. Respiratory tract lesions in noninhalation studies. *Toxicologic Pathology* 35:170-177.

Serafini-Cessi, F. 1972. Conversion of allyl alcohol into acrolein by rat liver. *Biochem J* 128:1103-1107.

Shelby, M. D., Erexson, G. L., Hook, G. J., and Tice, R. R. 1993. Evaluation of a three-exposure mouse bone marrow micronucleus protocol: Results with 49 chemicals. *Environmental and molecular mutagenesis* 21:160-179.

Shelby, M. D., and Witt, K. L. 1995. Comparison of results from mouse bone marrow chromosome aberration and micronucleus tests. *Environmental and molecular mutagenesis* 25:302-313.

Sinsheimer, J. E., Chen, R., Das, S. K., Hooberman, B. H., Osorio, S., and You, Z. 1993. The genotoxicity of enantiomeric aliphatic epoxides. *Mutat Res* 298:197-206.

Sisk, S. C., Pluta, L. J., Meyer, K. G., Wong, B. C., and Recio, L. 1997. Assessment of the in vivo mutagenicity of ethylene oxide in the tissues of B6C3F1 lacI transgenic mice following inhalation exposure. *Mutat Res* 391:153-164.

Solangi, K., Sacerdoti, D., Goodman, A. I., Schwartzman, M. L., Abraham, N. G., and Levere, R. D. 1988. Differential effects of partial hepatectomy on hepatic and renal heme and cytochrome P450 metabolism. *Am J Med Sci* 296:387-391.

Solt, D. B., Medline, A., and Farber, E. 1977. Rapid emergence of carcinogen-induced hyperplastic lesions in a new model for the sequential analysis of liver carcinogenesis. *American Journal of Pathology* 88:595-618.

Stott, W. T., and Gollapudi, B. B. 2001. Chapter 85 - 1,3-Dichloropropene. In *Handbook of Pesticide Toxicology (Second Edition)*, edited by R. I. Krieger, and W. C. Krieger. San Diego: Academic Press, pp. 1849-1859.

Stott, W. T., and Gollapudi, B. B. 2010. Chapter 106 - 1,3-Dichloropropene. In *Hayes' Handbook of Pesticide Toxicology (Third Edition)*, edited by R. Krieger. New York: Academic Press, pp. 2281-2292.

Stott, W. T., Johnson, K. A., Calhoun, L. L., Weiss, S. K., and Frauson, L. E. 1987. Telone* II Soil Fumigant: 2-year inhalation chronic toxicity-oncogenicity study in mice. (DPR Vol. No. 50046-0029, Record No. 060675).

Stott, W. T., Johnson, K. A., Jeffries, T. K., Haut, K. T., and Shabrang, S. N. 1995. Telone II soil fumigant: two-year chronic toxicity / oncogenicity study in Fischer 344 rats. Dow Chemical Company. Laboratory study ID #M-003993-031. (DPR Vol. No. 50046-098, Record No. 140562).

Stott, W. T., and Kastl, P. E. 1986. Inhalation pharmacokinetics of technical grade 1,3-dichloropropene in rats. *Toxicology and applied pharmacology* 85:332-341.

Stott, W. T., Miller, T. J., and Wardynski, A. K. 1997. 1,3-Dichloropropene: in vitro DNA binding. (DPR Vol. No. 50046-119, Record No. 162470).

Stott, W. T., Young, J. T., and Calhoun, L. L. 1984. Telone II soil fumigant: a 13-week inhalation study in rats and mice. (DPR Vol. No. 50046-038, Record No. 71713).

Streeter, C., Battjes, J., and Lomax, L. 1987. Telone II soil fumigant: an acute vapor inhalation study in Fischer 344 rats. (DPR Vol. No. 50046-032, Record No. 62070).

Tao, K. S., Urlando, C., and Heddle, J. A. 1993. Mutagenicity of methyl methanesulfonate (MMS) in vivo at the Dlb-1 native locus and a lacI transgene. *Environmental and molecular mutagenesis* 22:293-296.

Tates, A. D., Neuteboom, I., and Hofker, M. 1980. A micronucleus technique for detecting clastogenic effects of mutagens/carcinogens (DEN, DMN) in hepatocytes of rat liver in vivo. *Mutat Res* 74:11-20.

USEPA. 1994. Methods for Derivation of Inhalation Reference Concentrations and Application of Inhalation Dosimetry. (October 1994). http://www2.epa.gov/osa/methods-derivation-inhalation-reference-concentrations-and-application-inhalation-dosimetry.

USEPA. 1998. Reregistration Eligibility Decision (RED): 1,3-Dichloropropene. http://www.epa.gov/pesticides/reregistration/telone/.

USEPA. 2000. Benchmark Dose Technical Guidance Document.

USEPA 2002. A Review of the Reference Dose and Reference Concentration Processes. U.S. Environmental Protection Agency, Risk Assessment Forum, Washington, DC, EPA/630/P-02/002F, 2002.

USEPA 2004. Exposure Assessment Approaches For Chemicals Used As Soil Fumigants Consideration Of The Soil Fumigant Exposure Assessment System (SOFEA) - A Case Study With 1,3-Dichloropropene, pp. 21.

USEPA. 2007. Human health risk assessment: 1,3-dichloropropene. (authors: F. Fort, C. Olinger, E. Mendez and D. Vogel) *Office of Pesticide Programs, Health Effects Division, United States Environmental Protection Agency*.

USEPA. 2008a. Health Effects Support Document for 1,3-Dichloropropene 822-R-08-008. 142. http://www2.epa.gov/ccl/regulatory-determination-2-support-documents-13-dichloropropene-telone.

USEPA. 2008b. Updated RED Fact Sheet: 1, 3-Dichloropropene (1,3-D) http://www.epa.gov/pesticides/reregistration/REDs/factsheets/1,3-dichloropropene_fs.htm.

USEPA 2012a. Advances in inhalation gas dosimetry for derivation of a reference concentration (RfC) and use in risk assessment. United States Environmental Protection Agency, Washington, DC.

USEPA. 2012b. Benchmark Dose Technical Guidance Document. (June 2012).

Valencia, R., Mason, J. M., Woodruff, R. C., and Zimmering, S. 1985. Chemical mutagenesis testing in Drosophila. III. Results of 48 coded compounds tested for the national toxicology program. *Environmental Mutagenesis* 7:325-348.

Van Duuren, B. L., Goldschmidt, B. M., Loewengart, G., Smith, A. C., Melchionne, S., Seidman, I., and Roth, D. 1979. Carcinogenicity of halogenated olefinic and aliphatic hydrocarbons in mice. *Journal of the National Cancer Institute* 65:1433-1439.

van Wesenbeeck, I., and Cryer, S. 2014. Addendum to "Validation ofSOFEA2 with 1,3-Dichloropropene ambient monitoring data in Merced County California" describing Mixing Height adjustments. Regulatocy Science and Government Affairs, Dow AgroSciences LLC, 9330 Zionsville Road, Indianapolis, Indiana 46268-1: Dow AgroSciences LLC. (DPR Vol. No. 50046-0218, Record No. 282234) 58.

van Wesenbeeck, I., Cryer, S. A., and Helle, O. d. C. 2014. Validation Of SOFEA2 With 1,3-Dichloropropene Ambient Monitoring Data In Merced County California - Revised Report Dow Agrosciences Indianapolis, IN (5796) Dow Agrosciences LLC (DPR Vol. No. 50046-0210 Record No. 276639) 109.

Van Wesenbeeck, I., and Phillips, A. M. 2000. Field volatility of 1,3-dichloropropene and chloropicrin from surface drip irrigation application of In-Line to vegetables beds under polyethylene tarp. Indianapolis, Indiana 46268-1054: Dow AgroSciences LLC 990072. (DPR Vol. No. 50046-152, Record No. 178246).

van Wesenbeeck, I. J., and Cryer, S. A. 2004. SOFEA. SOil Fumigant Exposure Assessment system User's Guide. 67.

van Wesenbeeck, I. J., Cryer, S. A., and de Cirugeda Helle, O. 2015. Validation of SOFEA2 with 1,3-Dich1oropropene ambient monitoring data in Merced County California including mixing height modifications and border township effects. Regulatory Science and Government Affairs, Dow AgroSciences LLC, 9330 Zionsville Road, Indianapolis, Indiana 46268-1054. (DPR Vol. No. 50046-0230, Record No. 286007) 46.

van Wesenbeeck, I. J., Cryer, S. A., Havens, P. L., and Houtman, B. A. 2011. Use of SOFEA to predict 1,3-D concentrations in air in high-use regions of California. *Journal of environmental quality* 40:1462-1469.

Verder-Carlos, M. N. 2014. Recommendation on Township Cap Exception Requests for 1,3-Dichloropropene. Leahy, Brian R. , Reardon, Chris Department of Pesticide Regulation, from Verder-Carlos, Marylou N., Assistant Director. http://www.cdpr.ca.gov/docs/emon/methbrom/telone/rec_on_twnshp_telone.pdf.

Vos, R. M., van Welie, R. T., Peters, W. H., Evelo, C. T., Boogaards, J. J., Vermeulen, N. P., and van Bladeren, P. J. 1991. Genetic deficiency of human class mu glutathione S-transferase isoenzymes in relation to the urinary excretion of the mercapturic acids of Z- and E-1,3-dichloropropene. *Arch Toxicol* 65:95-99.

Waechter, J. M., Brzak, K. A., McCarty, L. P., LaPack, M. A., and Brownson, P. J. 1992. 1,3-Dichloropropene (Telone II Soil Fumigant) inhalation pharmacokinetics and metabolism in human volunteers. . (DPR Vol. No. 50046-052, Record No. 113124).

Waechter, J. M., and Kastl, P. E. 1988. 1,3-Dichloropropene: Pharmacokinetics and metabolism in Fischer 344 rats following repeated oral administration. (DPR Vol. No. 50046-0216, Record No. 282090).

Wahnschaffe, U., Bitsch, A., Kielhorn, J., and Mangelsdorf, I. 2005. Mutagenicity testing with transgenic mice. Part I: Comparison with the mouse bone marrow micronucleus test. *J Carcinog [serial online journal]* 4:3

Watson, W. P., Brooks, T. M., Huckle, K. R., Hutson, D. H., Lang, K. L., Smith, R. J., and Wright, A. S. 1987. Microbial mutagenicity studies with (Z)-1,3-dichloropropene. *Chem Biol Interact* 61:17-30.

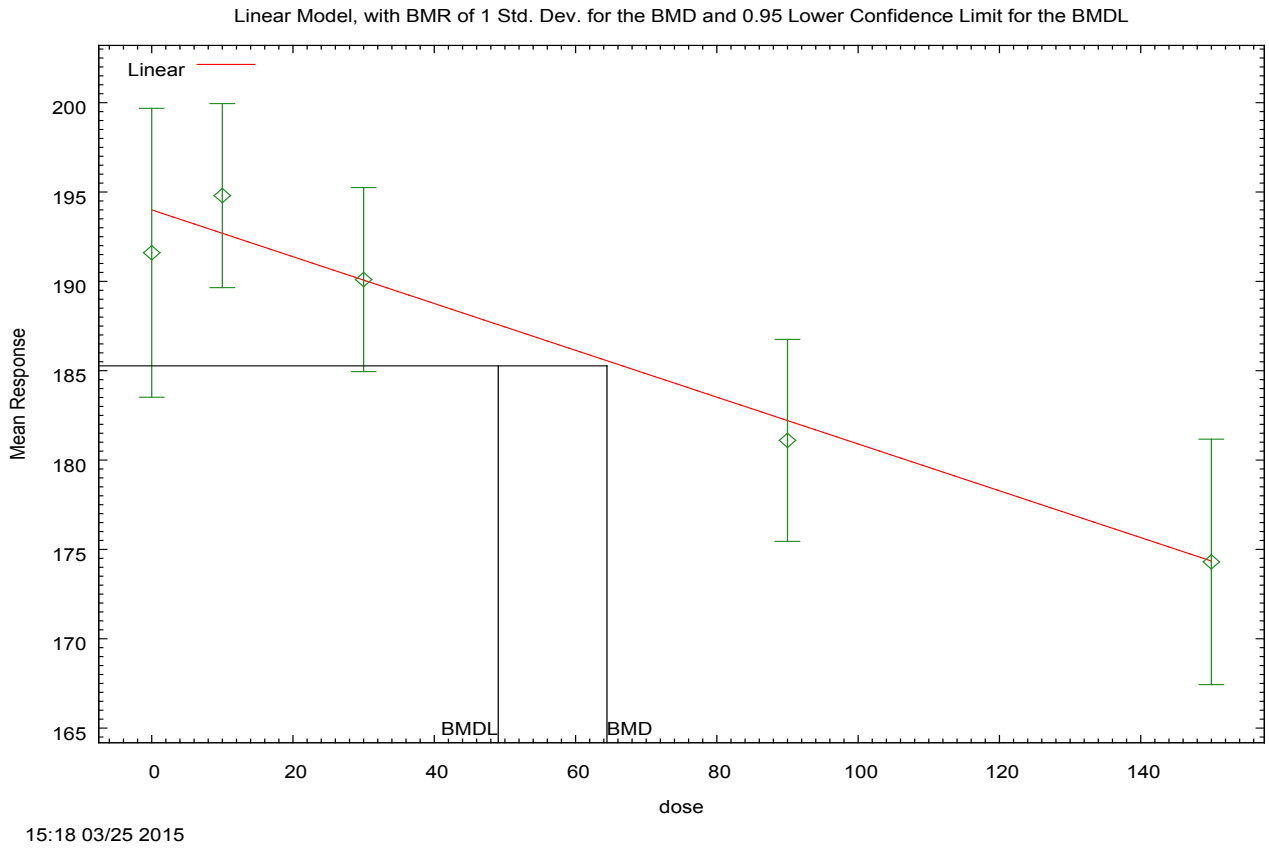
Wiley, J. A. 1991. *Study of children's activity patterns : final report, contract no. A733-149*. Sacramento, CA: California Environmental Protection Agency, Air Resources Board, Research Division.

Wiley, J. A., California Environmental Protection Agency. Air Resources Board. Research Division., and University of California Berkeley. Survey Research Center. 1991. Activity patterns of California residents : final report contract no. A6-177-33, pp. 63, 73 p. The Division, Sacramento, Calif.

Wofford, P., Segawa, R., and Schreider, J. 2009. Pesticide Air Monitoring in Parlier, CA, pp. 92. California Environmental Protection Agency, Department of Pesticide Regulation.

Würgler, F. E., Sobels, F. H., and Vogel, E. 1984. Drosophila as an assay system for detecting genetic changes. In *Handbook of Mutagenicity Test Procedures, 2nd Edition*, edited by M. L. B.J. Kilbey, W. Nichols and C. Ramel. New York: Elsevier, pp. 555-601.

APPENDIX I. BENCHMARK CONCENTRATION ANALYSIS OF BODY WEIGHT DECREMENTS RESULTING FROM SHORT TERM EXPOSURES TO 1,3-D (STOTT *ET AL.*, 1984)




```
 ======================================================================
        Polynomial Model. (Version: 2.20;  Date: 10/22/2014)
        Input Data File: R:/PDF/Telone/BMD files/Telone Male Rats Subchronic
50046_038/lin_Telone Male Rat BW Subchronic 50046_038_Opt.(d)
        Gnuplot Plotting File:  R:/PDF/Telone/BMD files/Telone Male Rats Subchronic
50046_038/lin_Telone Male Rat BW Subchronic 50046_038_Opt.plt
                                              Wed Jul 29 11:52:41 2015
 ======================================================================

 BMDS Model Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the response function is:

   Y[dose] = beta_0 + beta_1*dose + beta_2*dose^2 + ...


   Dependent variable = Mean
   Independent variable = Dose
   rho is set to 0
   Signs of the polynomial coefficients are not restricted
   A constant variance model is fit
```

Total number of dose groups = 5
Total number of records with missing values = 0
Maximum number of iterations = 500
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial Parameter Values
alpha = 77.188
rho = 0 Specified
beta_0 = 193.737
beta_1 = -0.131369

Asymptotic Correlation Matrix of Parameter Estimates

(*** The model parameter(s) -rho
have been estimated at a boundary point, or have been specified by the user,
and do not appear in the correlation matrix)

	alpha	beta_0	beta_1
alpha	1	-3.3e-007	-7.2e-010
beta_0	-3.3e-007	1	-0.7
beta_1	-7.2e-010	-0.7	1

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
alpha	71.6778	14.3356	43.5805	99.775
beta_0	193.737	1.68686	190.43	197.043
beta_1	-0.131369	0.0212188	-0.172957	-0.0897813

Table of Data and Estimated Values of Interest

Dose	N	Obs Mean	Est Mean	Obs Std Dev	Est Std Dev	Scaled Res.
0	10	192	194	11.3	8.47	-0.798
10	10	195	192	7.2	8.47	0.888
30	10	190	190	7.2	8.47	0.114
90	10	181	182	7.9	8.47	-0.304
150	10	174	174	9.6	8.47	0.1

Model Descriptions for likelihoods calculated

Model A1: Yij = Mu(i) + e(ij)
Var{e(Ij)} = Sigma^2

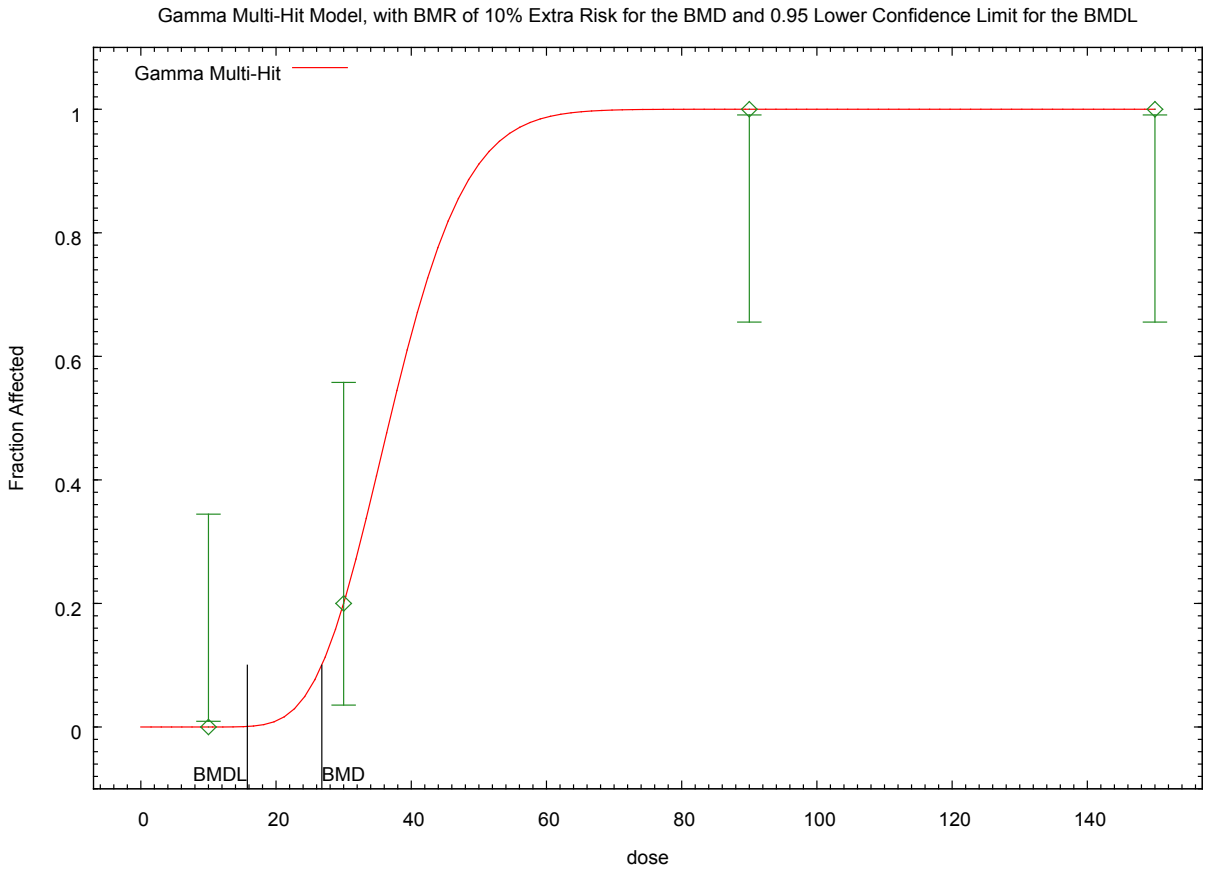
```
 Model A2:        Yij = Mu(i) + e(ij)
          Var{e(ij)} = Sigma(i)^2

 Model A3:        Yij = Mu(i) + e(ij)
          Var{e(ij)} = Sigma^2
     Model A3 uses any fixed variance parameters that
     were specified by the user

 Model  R:         Yi = Mu + e(i)
           Var{e(i)} = Sigma^2


                      Likelihoods of Interest

            Model      Log(likelihood)   # Param's      AIC
             A1         -131.022087            6     274.044174
             A2         -129.381893           10     278.763787
             A3         -131.022087            6     274.044174
          fitted        -131.804467            3     269.608935
              R         -146.031104            2     296.062209


                   Explanation of Tests

 Test 1:  Do responses and/or variances differ among Dose levels?
          (A2 vs. R)
 Test 2:  Are Variances Homogeneous? (A1 vs A2)
 Test 3:  Are variances adequately modeled? (A2 vs. A3)
 Test 4:  Does the Model for the Mean Fit? (A3 vs. fitted)
 (Note:  When rho=0 the results of Test 3 and Test 2 will be the same.)

                     Tests of Interest

   Test    -2*log(Likelihood Ratio)  Test df        p-value

   Test 1              33.2984          8          <.0001
   Test 2              3.28039          4           0.512
   Test 3              3.28039          4           0.512
   Test 4              1.56476          3          0.6674

The p-value for Test 1 is less than .05.  There appears to be a
difference between response and/or variances among the dose levels
It seems appropriate to model the data

The p-value for Test 2 is greater than .1.  A homogeneous variance
model appears to be appropriate here


The p-value for Test 3 is greater than .1.  The modeled variance appears
 to be appropriate here

The p-value for Test 4 is greater than .1.  The model chosen seems
to adequately describe the data


              Benchmark Dose Computation

Specified effect =            1

Risk Type        =     Estimated standard deviations from the control mean

Confidence level =         0.95
```

```
  BMD =        64.4463
BMDL =        49.0566
```

APPENDIX II. MODEL FAMILY PARAMETERS FROM BENCHMARK CONCENTRATION ANALYSIS OF BODY WEIGHT DECREMENTS RESULTING FROM SHORT TERM EXPOSURES TO 1,3-D

Model Name	BMR	BMD	BMDL	p-value Test 1: Lack dose response?	p-value Test 2: Constant variance?	p-value Test 3: Good variance model?	p-value for fit: Does the model for the mean fit?	AIC	Scaled residual for dose group nearest the BMD	Scaled residual for control group
Exponential2	1 SD	62.553	47.0028	< 0.0001	0.512	0.512	0.6655	269.6172	-0.2478	-0.832
Exponential3	1 SD	69.7925	47.2939	< 0.0001	0.512	0.512	0.4862	271.4864	-0.4017	-0.6679
Exponential4	1 SD	62.553	47.0028	< 0.0001	0.512	0.512	0.4554	271.6172	-0.2478	-0.832
Exponential5	1 SD	67.7718	35.4638	< 0.0001	0.512	0.512	0.3226	273.0227	0.04652	-0.5562
Hill	1 SD	66.5911	34.8983	<.0001	0.512	0.512	0.3284	272.999213	0.0625	-0.558
Linear	1 SD	64.4463	49.0566	<.0001	0.512	0.512	0.6674	269.608935	-0.304	-0.798
Polynomial	1 SD	64.5034	34.587	<.0001	0.512	0.512	0.4573	271.608929	-0.306	-0.797
Polynomial	1 SD	71.94	33.4165	<.0001	0.512	0.512	0.3181	273.040863	0.0682	-0.437

APPENDIX III. BENCHMARK CONCENTRATION ANALYSIS OF NASAL EPITHELIAL HYPERPLASIA RESULTING FROM SUBCHRONIC EXPOSURES TO 1,3-D (STOTT *ET AL.*, 1984)




```
================================================================
        Gamma Model. (Version: 2.16;  Date: 2/28/2013)
        Input Data File: R:/PDF/Telone/BMD files/Subchronic BMD/Telone Subchronic BMD
Files/gam_Telone SubC Stott 1984 Rat M RespHyper All Edit_Opt.(d)
        Gnuplot Plotting File:  R:/PDF/Telone/BMD files/Subchronic BMD/Telone
Subchronic BMD Files/gam_Telone SubC Stott 1984 Rat M RespHyper All Edit_Opt.plt
                                                Wed Dec 16 16:58:51 2015
 ================================================================

 BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response]= background+(1-background)*CumGamma[slope*dose,power],
   where CumGamma(.) is the cummulative Gamma distribution function


   Dependent variable = Effect
   Independent variable = Dose
   Power parameter is restricted as power >=1
```

Total number of observations = 4
Total number of records with missing values = 0
Maximum number of iterations = 500
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial (and Specified) Parameter Values
Background = 0.0416667
Slope = 0.0229063
Power = 1.5945

Asymptotic Correlation Matrix of Parameter Estimates

(*** The model parameter(s) -Background -Power
have been estimated at a boundary point, or have been specified by the user,
and do not appear in the correlation matrix)

	Slope
Slope	1

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
Background	0	NA		
Slope	0.478922	0.0549712	0.371181	0.586664
Power	18	NA		

NA - Indicates that this parameter has hit a bound
implied by some inequality constraint and thus
has no standard error.

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-5.00402	4			
Fitted model	-5.00411	1	0.000166762	3	1
Reduced model	-27.5256	1	45.0431	3	<.0001

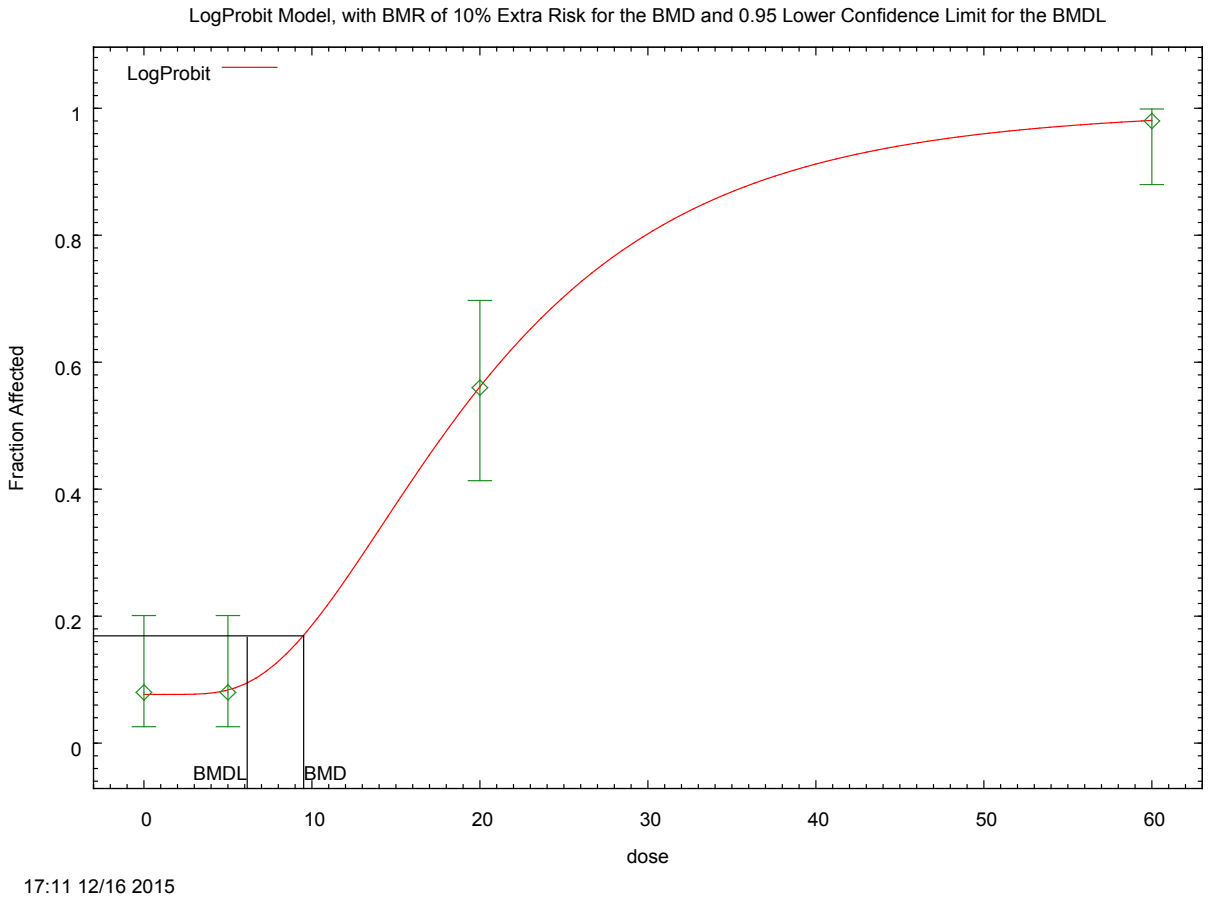
AIC: 12.0082

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
10.0000	0.0000	0.000	0.000	10.000	-0.006
30.0000	0.2000	2.000	2.000	10.000	-0.000
90.0000	1.0000	10.000	10.000	10.000	0.007
150.0000	1.0000	10.000	10.000	10.000	0.000

```
 Chi^2 = 0.00      d.f. = 3        P-value = 1.0000

   Benchmark Dose Computation

Specified effect =            0.1

Risk Type        =      Extra risk

Confidence level =           0.95

             BMD =        26.7719

            BMDL =       15.7543
```

APPENDIX IV. BENCHMARK CONCENTRATION ANALYSIS OF NASAL EPITHELIAL HYPERPLASIA RESULTING FROM CHRONIC EXPOSURES TO 1,3-D (STOTT *ET AL.*, 1987)




```
=====================================================================
        Probit Model. (Version: 3.3;  Date: 2/28/2013)
        Input Data File: R:/PDF/Telone/BMD files/Chronic BMD/Telone Chronic BMD
files/lnp_Stott 87 Mouse F U Nasal Resp Epi HyperTr All_Opt.(d)
        Gnuplot Plotting File:  R:/PDF/Telone/BMD files/Chronic BMD/Telone Chronic
BMD files/lnp_Stott 87 Mouse F U Nasal Resp Epi HyperTr All_Opt.plt
                                              Wed Dec 16 17:11:45 2015
 =====================================================================

 BMDS_Model_Run
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   The form of the probability function is:

   P[response] = Background
               + (1-Background) * CumNorm(Intercept+Slope*Log(Dose)),

   where CumNorm(.) is the cumulative normal distribution function


   Dependent variable = Effect
   Independent variable = Dose
   Slope parameter is not restricted
```

Total number of observations = 4
Total number of records with missing values = 0
Maximum number of iterations = 500
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

User has chosen the log transformed model

Default Initial (and Specified) Parameter Values
background = 0.08
intercept = -5.16282
slope = 1.75048

Asymptotic Correlation Matrix of Parameter Estimates

	background	intercept	slope
background	1	-0.27	0.24
intercept	-0.27	1	-0.99
slope	0.24	-0.99	1

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
background	0.0768112	0.0283114	0.021322	0.1323
intercept	-5.33137	1.29655	-7.87255	-2.79018
slope	1.798	0.401885	1.01032	2.58568

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-67.0754	4			
Fitted model	-67.0838	3	0.0167486	1	0.897
Reduced model	-136.371	1	138.591	3	<.0001

AIC: 140.168

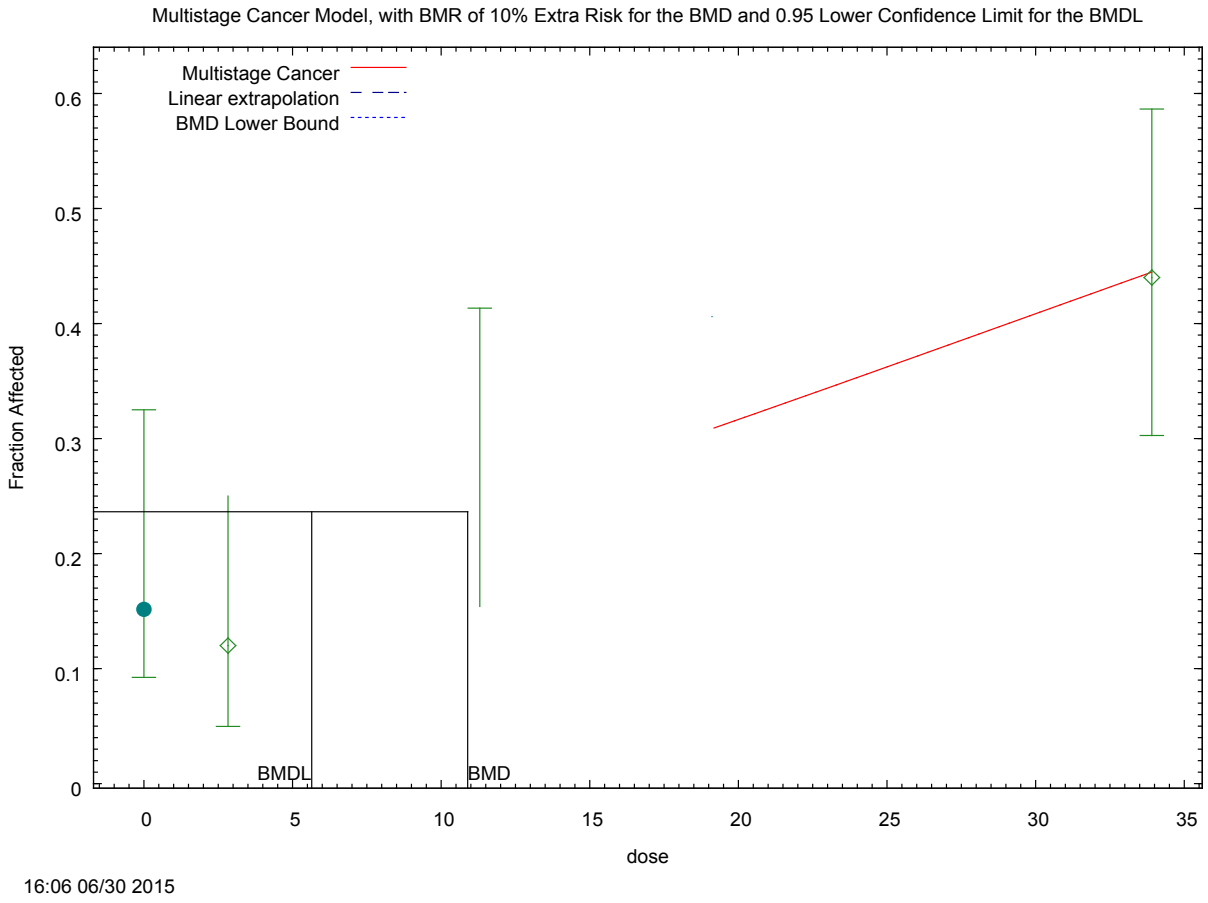
Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.0768	3.841	4.000	50.000	0.085
5.0000	0.0836	4.182	4.000	50.000	-0.093
20.0000	0.5586	27.932	28.000	50.000	0.019
60.0000	0.9805	49.023	49.000	50.000	-0.024

```
 Chi^2 = 0.02      d.f. = 1        P-value = 0.8971

   Benchmark Dose Computation

Specified effect =            0.1

Risk Type        =      Extra risk

Confidence level =           0.95

             BMD =        9.51048

            BMDL =        6.14742
```

APPENDIX V. QUANTITATIVE DOSE MODELING OF BRONCHIOLOALVEOLAR ADENOMA INCIDENCE IN MALE MICE FOLLOWING 2 YEARS OF DAILY EXPOSURE TO 1,3-D (STOTT *ET AL.*, 1987)

A. Resident / bystander / ambient analysis




```
 ======================================================================
        Multistage Model. (Version: 3.4;  Date: 05/02/2014)
        Input Data File: C:/USEPA/BMDS260/Data/msc_Male mouse pulmonary adenomas,
res-byst, Telone_Opt.(d)
        Gnuplot Plotting File:  C:/USEPA/BMDS260/Data/msc_Male mouse pulmonary
adenomas, res-byst, Telone_Opt.plt
                                                Tue Jun 30 16:06:51 2015
 ======================================================================

 BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(
                 -beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive
```

```
  Dependent variable = Incidence
  Independent variable = HEC-RB

Total number of observations = 4
Total number of records with missing values = 0
Total number of parameters in model = 3
Total number of specified parameters = 0
Degree of polynomial = 2


Maximum number of iterations = 500
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008


                 Default Initial Parameter Values
                    Background =     0.147811
                       Beta(1) =    0.0106136
                       Beta(2) =  5.3849e-005


          Asymptotic Correlation Matrix of Parameter Estimates

            Background      Beta(1)      Beta(2)

Background           1         -0.6         0.47

   Beta(1)         -0.6            1        -0.95

   Beta(2)         0.47        -0.95            1


                              Parameter Estimates

                                                      95.0% Wald Confidence
Interval
       Variable         Estimate        Std. Err.     Lower Conf. Limit   Upper Conf.
Limit
     Background         0.151509        0.0422438           0.0687127
0.234305
        Beta(1)       0.00834749        0.0121548          -0.0154754
0.0321704
        Beta(2)      0.000121749        0.0003686        -0.000600693
0.000844191


                        Analysis of Deviance Table

       Model      Log(likelihood)  # Param's  Deviance  Test d.f.   P-value
     Full model         -104.36         4
   Fitted model        -105.156         3        1.5927      1          0.2069
  Reduced model        -111.888         1       15.0568      3        0.001769

          AIC:         216.313
```

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.1515	7.424	9.000	49.000	0.628
2.8300	0.1721	8.606	6.000	50.000	-0.976
11.3000	0.2398	11.750	13.000	49.000	0.418
33.9100	0.4442	22.210	22.000	50.000	-0.060

Chi^2 = 1.53 d.f. = 1 P-value = 0.2167

Benchmark Dose Computation

Specified effect = 0.1

Risk Type = Extra risk

Confidence level = 0.95

BMD = 10.8916

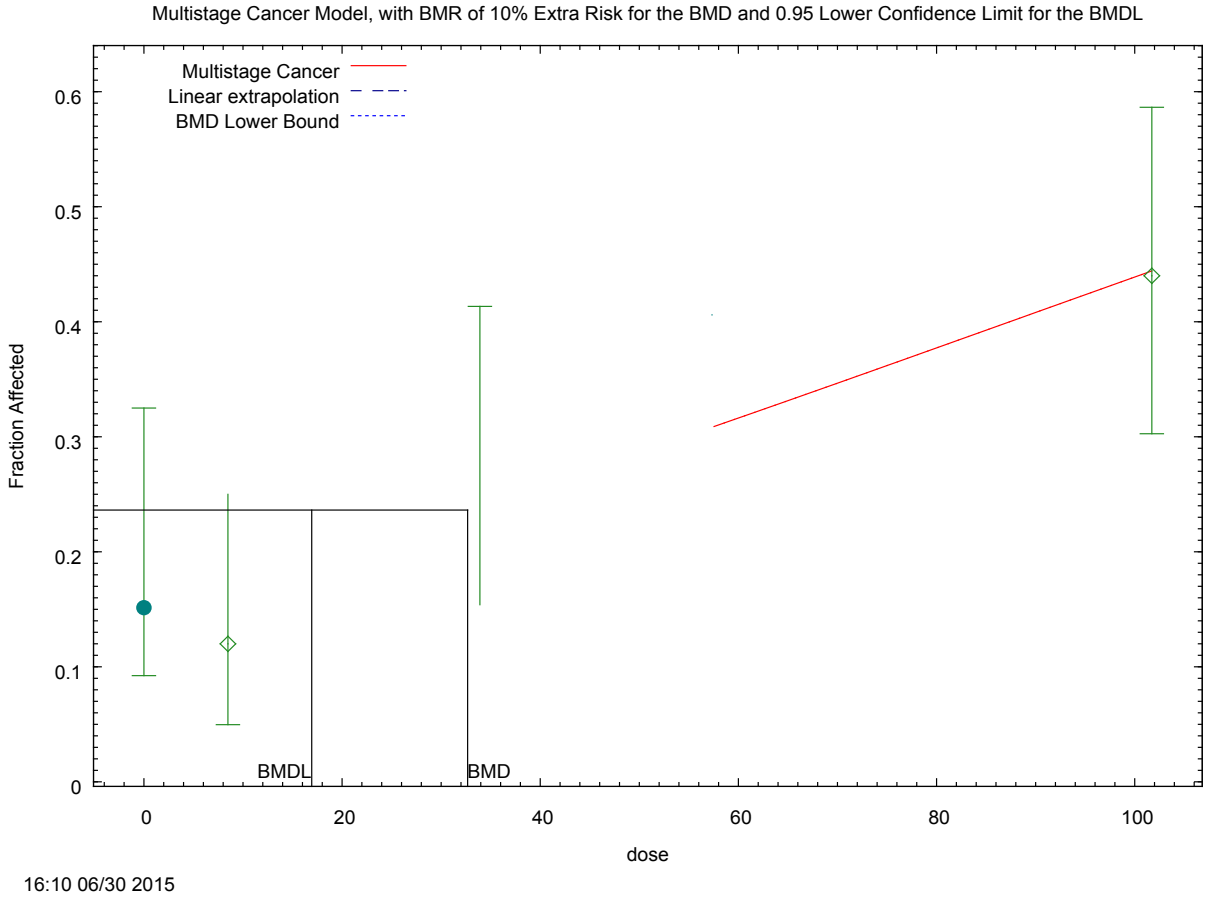
BMDL = 5.64544

BMDU = 23.2709

Taken together, (5.64544, 23.2709) is a 90 % two-sided confidence interval for the BMD

Cancer Slope Factor = 0.0177134

B. Occupational analysis




```
 ======================================================================
        Multistage Model. (Version: 3.4;  Date: 05/02/2014)
        Input Data File: C:/USEPA/BMDS260/Data/msc_Male mouse pulmonary adenomas,
res-byst, Telone_Opt.(d)
        Gnuplot Plotting File:  C:/USEPA/BMDS260/Data/msc_Male mouse pulmonary
adenomas, res-byst, Telone_Opt.plt
                                              Tue Jun 30 16:10:34 2015
 ======================================================================

 BMDS_Model_Run
~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~~

   The form of the probability function is:

   P[response] = background + (1-background)*[1-EXP(
                 -beta1*dose^1-beta2*dose^2)]

   The parameter betas are restricted to be positive


   Dependent variable = Incidence
   Independent variable = HEC-occ
```

Total number of observations = 4
Total number of records with missing values = 0
Total number of parameters in model = 3
Total number of specified parameters = 0
Degree of polynomial = 2

Maximum number of iterations = 500
Relative Function Convergence has been set to: 1e-008
Parameter Convergence has been set to: 1e-008

Default Initial Parameter Values
Background = 0.147811
Beta(1) = 0.00353787
Beta(2) = 5.98309e-006

Asymptotic Correlation Matrix of Parameter Estimates

	Background	Beta(1)	Beta(2)
Background	1	-0.6	0.47
Beta(1)	-0.6	1	-0.95
Beta(2)	0.47	-0.95	1

Parameter Estimates

Variable	Estimate	Std. Err.	95.0% Wald Confidence Interval Lower Conf. Limit	Upper Conf. Limit
Background	0.151502	0.0422361	0.0687212	0.234284
Beta(1)	0.00278329	0.00405143	-0.00515736	0.0107239
Beta(2)	1.35201e-005	4.09563e-005	-6.67528e-005	9.37931e-005

Analysis of Deviance Table

Model	Log(likelihood)	# Param's	Deviance	Test d.f.	P-value
Full model	-104.36	4			
Fitted model	-105.156	3	1.59141	1	0.2071
Reduced model	-111.888	1	15.0568	3	0.001769

AIC: 216.311

Goodness of Fit

Dose	Est._Prob.	Expected	Observed	Size	Scaled Residual
0.0000	0.1515	7.424	9.000	49.000	0.628
8.4800	0.1721	8.605	6.000	50.000	-0.976
33.9100	0.2398	11.752	13.000	49.000	0.418
101.7300	0.4442	22.210	22.000	50.000	-0.060

Chi^2 = 1.53 d.f. = 1 P-value = 0.2169

Benchmark Dose Computation

Specified effect = 0.1

Risk Type = Extra risk

Confidence level = 0.95

BMD = 32.67

BMDL = 16.9364

BMDU = 69.8076

Taken together, (16.9364, 69.8076) is a 90 % two-sided confidence interval for the BMD

Cancer Slope Factor = 0.00590445

APPENDIX VI. SUPPLEMENTAL DISCUSSION OF ISSUES CONCERNING THE GENOTOXICITY TESTING OF 1,3-D

Results for the genotoxicity testing of 1,3-D using various *in vitro* and *in vivo* assays are summarized in Tables III.11a and III.11b The former shows the studies submitted by the Registrant in the context of satisfying data requirements under SB950 while the latter shows the studies conducted by the National Toxicology Program for its Gene Tox Program in the 1980's. This supplement is meant to cover several things of importance to understanding the genotoxicity testing in the aforementioned tables as well as in a few studies found in the open literature that were not included in these tables. This section will begin with a discussion of how test conditions are known to affect testing with the *Salmonella* tester strains. This is followed by synopses of the testing done by the National Toxicology Program for its Gene Tox Program (except for the bacterial testing since the testing and results are sufficiently addressed by Table III.11b). The next section will discuss the following publications from the open literature concerning the *in vivo* testing in rats using the alkaline-elution assay for detection of DNA damage (Ghia *et al.*, 1993; Kitchin *et al.*, 1993; Kitchin and Brown, 1994). The final section will discuss submissions from the Registrant indicating a lack of mutagenicity in testing using transgenic (Big Blue) mice (Gollapudi and Cieszlak, 1997). In Appendix VII the genotoxicity testing of 1,3-D metabolites and degradates is discussed.

I. Testing with the *Salmonella* tester strains for induction of gene mutations (Ames Test)

1,3-D is mutagenic in the Ames Test without or with the use of metabolic-activation systems. The potency of the mutagenic response is affected significantly by the presence of autoxidation products in the test material as well as the conduct of the testing.

Air-exposed test material. 1,3-D (as a single isomer [*cis* or *trans*] or a mixture of isomers) that has been obtained from a chemical supply company without purification by silica-gel, column chromatography before testing typically is directly mutagenic in the Ames Test. For example, using the *Salmonella typhimurium* tester strain TA100 in the standard plate-incorporation assay, without any metabolic-activation system, an unpurified 1,3-D isomer mixture induced 97 revertants/µmole whereas after passage through a silica gel column, only 13 revertants/µmole were induced (Eder *et al.*, 2006) The mutagenic impurities removed by the silica-gel treatment are assumed to be autoxidation products given that when the purified 1,3-D isomer mixture was stored for 10 weeks at 20ºC under air then tested as before against TA100, the mutagenicity of the test material increased to77 revertants/µmole but if the storage was for 10 weeks at 4ºC under argon, there was no such increase (16 revertants/µmole measured). The mutagenic impurities removed by the silica-gel treatment include 1,3-dichloro-1,2-epoxypropane (hereafter referred to as 1,3-D

epoxide), 2,3-dichloropropanal, and 2-chloroacrolein (Watson *et al.*, 1987; Eder *et al.*, 2006). It should be underscored that these impurities are not removed by distillation or preparative gas chromatography, despite achieving very high purity levels with these procedures (Watson *et al.*, 1987; Eder *et al.*, 2006). Watson *et al.* (1987) have proposed that autoxidation begins with radical formation on carbon 3 of 1,3-D as chlorine radical is lost, followed by reaction with oxygen to give an alkyl peroxy radical, which in turn reacts with another 1,3-D molecule to produce eventually both isomers of 1,3-D epoxide. Eder *et al.* (2006) have proposed that the 1,3-D epoxides formed in the test material readily rearrange to 2,3-dichloropropanal, which can dehydrochlorinate to produce 2-chloroacrolein. Although these autoxidation products are present at low concentrations in technical materials (<1%), they are potent, direct-acting mutagens in TA100. 2-Chloroacrolein induced 150,000 revertants/µmole using the standard plate incorporation assay and no metabolic activation system while under these same test conditions, the *cis* and *trans* isomers of 1,3-D epoxide induced 37,000 and 17,000 revertants/µmole, respectively (Schneider *et al.*, 1998a).

That the commonly used solvent dimethyl sulfoxide (DMSO) significantly may affect the mutagenicity testing of 1,3-D containing autoxidation products is important. As noted by Watson *et al.* (1987) and explored further by Schneider *et al.*(1998a), 1,3-D epoxide (either isomer) dissolved in DMSO is quantitatively converted to 2,3-dichloropropanal, which in turn undergoes loss of HCl to form 2-chloroacrolein. Ten minutes after addition of 1,3-D (mixture of isomers) to DMSO at room temperature, 2,3-dichloropropanal can be detected. After 155 minutes, no 1,3-D epoxide is left; rather the DMSO solution now consists only of 2,3-dichloropropanal and 2-chloroacrolein in equal amounts. After 280 minutes, the only chemical detected in the DMSO solution is 2-chloroacrolein.

Plate incorporation assay *versus* preincubation assay. The potency of the mutagenicity of 1,3-D towards TA100 in testing not employing a metabolic-activation system is many folds greater when the test material and the tester bacteria are incubated together before molten agar is added and the contents are poured on the agar plate. Testing the same unpurified 1,3-D isomer mixture that gave 97 revertants/µmole with the standard plate assay, Eder *et al*. (2006) observed that using a 30-minute preincubation period increased the mutagenicity to 424 revertants/µmole. In the case of testing unpurified, 95% *trans*-1,3-D material, the difference was 10 fold: 574 revertants/µmole were measured upon using a 30-minute preincubation period, compared to 57 revertants/µmole from following the standard plate-incorporation assay. Possible reasons for the large differences in mutagenic potency based on assay type include the following: for the duration of the preincubation step, the bacteria are exposed to higher test concentrations than would occur in the standard plate-incorporation assay; and the preincubation limits the escape of volatile chemi-

cals, thus increasing the chances that these chemicals will undergo chemical reactions, including DNA adduct formation.

Amount of S9 and duration of the preincubation period. 1,3-D is activated by metabolic activation systems like liver microsomes (Watson *et al.*, 1987; Schneider *et al.*, 1998a) and S9 (Neudecker and Henschler, 1986). In the testing with S9, which was done using a preincubation step, the mutagenic potency was dependent on the concentration of the S9 used for metabolic activation as well as the length of time that the S9 mix, chemical and tester bacteria were together before molten agar was added and the contents were poured onto agar plates. Neudecker and Henschler (1986) tested *cis*-1,3-D (>99% purity after preparative gas chromatography) using TA100 and S9 mix prepared from liver from Aroclor-1254-induced male Wistar rats. Two protein concentrations for the S9, 4 *versus* 12 mg/mL of S9 mix, and two preincubation durations, 20 *versus* 120 minutes, were compared. During the preincubation period, the incubation volumes were sealed under air-tight caps and shaken in a water bath at 37ºC. Using a 20-minute preincubation, 440 revertants/µmole were induced regardless of which protein concentration for the S9 was used. By contrast, with a 120-minute preincubation, 1740 and 3040 revertants/µmole were observed using the lower and higher protein concentrations for the S9, respectively.

1,3-D free of autoxidation products is still an alkylating agent. As already noted, the mutagenic potency of 1,3-D towards TA100 without S9 activation is decreased after it has been passed through a column of silica gel. The fact that some direct mutagenicity remains even though the autoxidation products are undetectable is consistent with the supposition that since the chlorine on the allylic carbon (carbon 3) can serve as a leaving group in nucleophilic substitution reactions, 1,3-D should be a direct-acting, alkylating agent (Creedy *et al.*, 1984). Two lines of experimentation support this supposition.

First, if 1,3-D is incubated at 100ºC for 10 minutes with the model nucleophile, 4-(4-nitrobenzyl)pyridine (NBP), subsequent addition of triethylamine produces a violet color (Eder *et al.*, 1982a; Eder *et al.*, 1982b; Eder *et al.*, 2006). [17] The characteristic color development was observed when testing a mixture of the isomers as well as when testing the isomers separately; also, comparable color intensity was measured when testing the different test materials before as well as after they were purified by passage through a column of silica gel (Eder *et al.*, 2006). The significance of the color development is that it only occurs if there has been adduct formation involving NBP's pyridine nitrogen. Thus, this so called "NBP test" constitutes a simple, colorimetric test for identifying alkylating agents (Epstein *et al.*, 1955). Furthermore, under identical

[17] The nonaqueous version of this assay was employed, *i.e.*, 1,3-D, NBP and triethylamine were dissolved in a nonaqueous solvent.

assay conditions and test concentrations, there is a quantitative relationship between the change in absorbance measured at 560 nm and alkylating potency (Eder *et al.*, 1980). Consequently, since 1,3-D gave increases in absorbance of 1.93-2.24 [18] (when tested as mixed isomers or as individual isomer, whether before or after silica gel treatment), 1,3-D can be considered to be a stronger alkylating agent than the following alkenes also having a chlorinated allylic carbon because the latter in the NBP test gave changes in absorbance (shown in parentheses) that were smaller: 3-chloro-1-propene [19] (0.29), 3-chloro-2-methyl-1-propene (0.57) and 1-chloro-2-butene (1.04).

Second, 1,3-D reacts with the thiol group in glutathione (GSH) in the absence of any enzymes or catalysts. For example, Vos *et al*. (1991) prepared GSH-1,3-D conjugates by dripping an aqueous solution of GSH (0.3 mM, pH 9.5) into ethanol containing 5 mM 1,3-D (each isomer done separately); after the resulting solution had incubated overnight, the GSH conjugates were isolated and purified by HPLC. In that same study, the authors reported that nonenzymatic conjugation of 1,3-D with GSH occurred at different rates with the two isomers: 0.15 *versus* 0.60 nmol/min for the *trans* and *cis* isomers, respectively, using incubations of 20 minutes at 37 ºC and pH 7.2.

II. Nonbacterial Testing of 1,3-D in the National Toxicology Program

A. *In vitro* mammalian-cell assays

L5178Y thymidine kinase$^{+/-}$ -3.7.2C mouse lymphoma cell mutation assay. Ethanol, three concentrations of 1,3-D (2.5, 5 and 10 nL/mL treatment media [27, 54 and 108 µM]) and methyl methanesulfonate (MMS [positive control], at 5 nL/mL treatment media [59 µM]) were tested in each of the two trials that were conducted (Myhr *et al.*, 1991). Murine cells in treatment media (Fischer's growth medium with 5% v/v horse serum) were exposed to 1,3-D for 4 hours at 37ºC, using sealed tubes that were rolled. A dose-response for resistance to the cytostatic pyrimidine analogue 5-trifluorothymidine was induced by 1,3-D in both trials. At the lowest concentration tested, 27µM, the relative total growth (RTG) ranged from 49 to 82% (over the total of 6 cultures from the two trials) and the average mutant frequency (expressed as mutants per 10^6 cells) increased from 29 and 32 in the solvent controls to 42 and 83 in the first and second trials, respectively; both increases were statistically significant ($p \leq 0.05$). At 108 µM, the RTG ranged from 5 to 31% and the average mutant frequency was increased by factors of 6 and 8 relative to the

[18] Note that an absorbance of 2 means that the light transmittance at 560 nm was reduced to 1%.
[19] This is allyl chloride (so 1,3-D *without* a chlorine on the end of the double bond).

values measured in the respective solvent controls. Testing at 216 µM in the first trial and 162 µM in the second trial also was done. With the former, the RTG was decreased to 1-9% and the average mutant frequency was increased to 353; this was comparable to the mutant frequency of 367 induced by MMS (59 µM) when tested in the first trial. Excessive toxicity negated the testing at 162 µM in the second trial.

Colony sizing was performed using an electronic counter fitted with a colony-size discriminator. Sizing was done on the following: one culture from the solvent-control testing in each of the trials; and one culture from the testing at 216 µM (first trial) and at 108 µM (second trial). The mutant frequencies of large *versus* small colonies (expressed as mutants per 10^6 cells) in the solvent controls were comparable in a given trial: 15 *versus* 12 in the first trial; and 18 *versus* 22 in the second trial, respectively. Exposure to 1,3-D increased the frequency of large-colony-forming mutants by a factor of 5 in both trials, despite the differences in the concentrations tested. However, 1,3-D increased the frequency of small-colony forming mutants by fjactors of 15 and 13 in the testing of 216 µM (first trial) and 108 µM (second trial), respectively. The preferential induction of small-colony-forming mutants has this significance for this assay. Such activity is associated with chemicals that induce large changes in genetic material like chromosomal aberrations, as opposed to just point mutations or small deletions at the thymidine kinase locus (Moore and Doerr, 1990).

Sister-chromatid exchanges (SCE) in cultured Chinese hamster ovary (CHO) cells. In Loveday *et al.* (1989), CHO cells were exposed to 1,3-D without the use of a metabolic activation system in the first two trials, followed by a single trial using S9. In this testing, dilutions of 1,3-D were prepared using DMSO.

For the first two trials, cells in culture medium (McCoy's 5A modified [20] buffered with sodium bicarbonate [21]) were exposed for 2 hours at 37ºC to 1,3-D before 5-bromo-2'-deoxyuridine (BrdUrd) was added. The incubation of the cells with 1,3-D and now BrdUrd was continued for another 24 hours, thus giving the cells a total of 26 hours of exposure to 1,3-D. Afterwards, cells were rinsed and incubated in medium containing BrdUrd and colcemid for an additional 2 – 2.5 hours. When enumeration of first-division (M1) and second-division (M2) cells indicated that less than 80% of the cells were M2, the test chemical was considered to have caused cell-cycle delay. In such cases, the exposure to BrdUrd was extended by 4 hours, thus giving the cells a total of 30 – 30.5 hours of exposure to BrdUrd.

For the testing with S9 (third trial), cells were placed in serum-free medium containing S9 as well as its supporting cofactors, 1,3-D was added, and the resulting mixture was incubated at 37ºC for 2 hours. Afterwards, the cells were rinsed and incubated for 24 hours in medium containing BrdUrd. Subsequently, colcemid was added and the incubation was continued for another 2 – 2.5 hours. Thus, the testing with metabolic activation involved exposing the cells to 1,3-D for just 2 hours and to BrdUrd for 26 – 26.5 hours.

In the first trial, DMSO, 1,3-D (0.995, 9.95 and 29.9 µg/mL [9, 90 and 269 µM, respectively]) and mitomycin C (MMC; 0.0015 and 0.01 µg/mL [0.0135 and 0.090 µM, respectively]) were tested. At the low concentration, no increase in SCE's was observed relative to the 8.00 SCE's per cell observed with the DMSO control. However, at the mid and high concentrations, the numbers of SCE's per cell were 9.56 and 13.60; these constituted increases of 20% and 70%, respectively, over the DMSO control value. Apparently, because at the high concentration, cell-cycle delay was observed, all treatments groups in the first trial (including both MMC treatments) underwent a total of 30.5 hours of exposure to BrdUrd.

For the second trial, the 1,3-D test concentrations were changed to 30, 40 and 50 µg/mL (270, 360 and 451 µM, respectively). Only the high concentration resulted in cell-cycle delay (and underwent 30.5 hours of BrdUrd exposure). The numbers of SCE's per cell observed in the low, mid and high concentration were 13.32, 17.12 and 19.70, respectively *versus* 7.70 SCE's per cell in the DMSO control. For comparison, the numbers of SCE's per cell from testing MMC at 0.0135 and 0.090 µM were 11.30 and 29.90, respectively. Based on a linear regression (trend)

[20] It was not addressed whether 10% v/v fetal bovine serum (possibly containing glutathione), which was present for growing cells, was used for the SCE testing done without a metabolic-activation system. In the testing done with S9, it was indicated that the exposure to test chemicals occurred in serum-free medium.
[21] HEPES, an amine buffer, was used at 20 mM in place of bicarbonate for exposures involving "liquid, volatile chemicals in sealed flasks." Whether this was done in the testing of 1,3-D was not addressed.

test, the dose response from testing 1,3-D was statistically significant ($p < 0.005$ (Galloway *et al.*, 1987)).

For the third trial, DMSO, 1,3-D (2.99, 9.95 and 29.9 µg/mL [27, 90 and 269 µM, respectively]) and cyclophosphamide (0.5 and 2.5 µg/mL [1.8 and 9.0 µM, respectively) were tested. At the low concentration, there was no increase in SCE's relative to the 7.70 SCE's per cell observed with the DMSO control; but at the mid and high concentrations, the numbers of SCE's per cell were 10.00 and 13.20, respectively. For comparison, the numbers of SCE's per cell from testing cyclophosphamide at 1.8 and 9.0 µM were 15.00 and 35.80, respectively. Based on the trend test, the dose response from testing 1,3-D was statistically significant ($P < 0.005$ (Galloway *et al.*, 1987)).

Chromosome aberrations in cultured Chinese hamster ovary (CHO) cells. Loveday *et al.* (1989) also reported on the experiments with CHO cells, using the same cell culture medium, for the induction of chromosome aberrations. Like the SCE testing, there were three trials: the first two involved exposure to 1,3-D without the use of a metabolic activation system, followed by a single trial using S9. As in the SCE testing, dilutions of 1,3-D were prepared using DMSO.

For the first two trials, cells in culture medium containing serum were exposed to 1,3-D for 8 hours at 37ºC. Afterwards, cells were rinsed and incubated for an additional 2 – 2.5 hours in medium containing serum to which colcemid had been added. For the testing with S9, cells were placed in serum-free medium containing S9 as well as its supporting cofactors, 1,3-D was added, and the resulting mixture was incubated at 37ºC for 2 hours. Afterwards, the cells were rinsed and incubated for 8 hours in medium containing serum. Subsequently, colcemid was added and the incubation was continued for another 2 hours. Thus, the testing that used S9 involved exposing cells to 1,3-D for just 2 hours, but whether S9 was used or not, cells were allowed 10 – 10.5 hours of growth in culture medium that contained fetal bovine serum.

In the first trial, DMSO, 1,3-D (4.91, 9.8, 29.5 and 49.1 µg/mL [44.2, 88.5, 265 and 442 µM, respectively]) and MMC (5 µg/mL [15 µM]) were tested. The only noteworthy effect occurred at highest concentration tested: 16 of the 100 cells scored exhibited simple aberrations, with one cell also showing a complex aberration. For comparison, the percent of cells showing complex and (or) simple aberrations from exposure to MMC was 50% (based on 50 cells scored). When the testing without metabolic activation was repeated, using 1,3-D concentrations of 451, 676

and 901 µM, 50% decreases in cell confluency were induced by each concentration, but no increase in aberrations was observed.

For the third trial, DMSO, 1,3-D (10, 30 and 50 µg/mL [90, 270 and 451 µM, respectively]) and cyclophosphamide (50 µg/mL [179 µM]) were tested. The maximum effect occurred with the high concentration: out of 100 cells scored, 3 cells showed simple aberrations while another 2 cells showed complex aberrations. For comparison, the percent of cells showing complex and (or) simple aberrations from exposure to cyclophosphamide was 36% (based on 50 cells scored).

Since reproducible chromosome damage was not observed in the testing without S9 and the testing with S9 at a 1,3-D concentration greater than 50 µg/mL (not specified) resulted in significant cell-cycle delay (no metaphases recovered), 1,3-D was considered to be negative for inducing chromosomal damage in this assay. However, the authors did note that 1,3-D (without and with the use of S9, test concentrations not specified) caused 50-100% of the cells to have chromatid gaps. These changes were not scored as chromosomal aberrations for the purposes of this testing. Inspection of the reporting indicates that no similar comments about gaps being observed were made with regards to any of the 19 other chemicals that were tested.

B. Testing in *Drosophila* for induction of sex-linked recessive lethals (SLRL's) and reciprocal translocations (RT's)

1,3-D was tested for the induction of SLRL's as well as RT's using feeding as the exposure route.[22] Testing by injection was not tried, in accordance with the guidelines under which the NTP sponsored testing was conducted (Valencia *et al.*, 1985). 1,3-D was dissolved in 10% ethanol and that solution was diluted with 5% aqueous sucrose to achieve the test concentration of 5750 ppm.[23] This test concentration had been selected based on previously conducted range-finding testing (data not provided). Canton-S wild-type males were placed in vials having glass filters soaked with the aqueous sucrose containing 1,3-D at 0 or 5750 ppm. Males were transferred each day without etherization to vials containing freshly made 1,3-D-sucrose mixture on filters. Mortality was recorded at the time of the transfers. After a total of 72 hours of exposure, a final mortality count was made and survivors were mated, depending on the endpoint. With this exposure method, 5750 ppm 1,3-D increased mortality and sterility by 33% and 10%, respectively, over what were measured with the 0 ppm males.

For the SLRL testing, surviving males were mated individually to three harems of *Basc* virgin females to produce three broods over 7 days. The total number of lethals was 20 from a total of 6584 treated chromosomes tested (0.30%). By comparison, the total of lethals was 8 from a total of 6918 negative-control chromosomes tested (0.12%). The increase in SLRL's was statistically significant ($P < 0.04$).

For the RT testing, surviving males were placed 10 to a vial containing 20 untreated, virgin females carrying genetic markers (*bw;st*) and mated for three days. Afterwards, the males were removed and the females were transferred to fresh medium lacking yeast [24] at 3-4-day intervals for a period of three weeks to produce a total of 6 broods. For 1,3-D, 6955 genomes were tested and a single translocation, T(2;3), was recovered. Although the historical, negative-control incidence for RT's, pooled from the three participating laboratories, indicated that a "spontaneous" translocation was a rare event in this testing (2/95352 genomes [0.002%]), the occurrence of the one translocation in the 1,3-D testing was not statistically significant based on the conditional binomial response test ($P>0.05$). In general, at least two translocations were required in this study design in order to achieve statistical significance.

C. Chromosomal damage *in vivo*

[22] Work was done at Bowling Green State University.
[23] It was not stated in the report whether "ppm" referred to weight/volume or volume/volume.
[24] The omission of yeast prevents the development of the ovaries and egg deposition. As a result, sperm from the mating are retained in the female's storage organs (the spermathecae and ventral receptacle). By storing the sperm, more chromosomal breaks may become available, thereby increasing the chances for the formation of a translocation (Würgler *et al.*, 1984).

Bone-marrow chromosomal aberration test. 1,3-D was one of the 65 chemicals tested in this assay for the National Toxicology Program (NTP) regarding its Gene Tox Program. However, the data for 1,3-D were never published, even though the results for the testing were included in reportings that made overall comparisons of the results for the 65 test chemicals (e.g., (Shelby and Witt, 1995)). Consequently, the NTP Archives were contacted regarding the testing of 1,3-D and the description that follows is based on the information that subsequently was provided in response.

The methods for this assay were described in McFee (1989) and the statistical analysis of the data was described in Shelby and Witt (1995). Testing consisted of two trials, each time using 8 male B6C3F1 mice per dose, including the vehicle and positive controls. 1,3-D was administered as a single intraperitoneal injection. Animals were sacrificed 17 hours after injection in each trial; 2 hours before sacrifice, animals had received an intraperitoneal injection of colchicine to allow the collection of metaphases from bone-marrow cells. The scoring was done using metaphases known to be at their first post-treatment cell division based on their uniformly dark-stained chromosomes. A total of 50 metaphases were scored in a blind fashion, with two observers each scoring 25 cells.

In the first trial, the doses were 0 (corn oil), 50, 100 and 200 mg/kg while for the second trial, only 0 and 200 mg/kg were tested. For each trial, the positive control was 7,12-dimethylbenz[a]-anthracene (DMBA) given at 100 mg/kg. In the first trial, the mean percent of cells with aberrations increased from 2.00 in the vehicle controls to 3.25 and 5.75 in the 100 and 200 mg/kg groups, respectively. The overall trend test ($p = 0.0001$) and pairwise comparison for the highest dose tested ($p = 0.003$) were significant. By comparison, the mean percent of aberrant cells for the DMBA group was 14.5%. The second trial confirmed the reproducibility of 1,3-D's effect: the mean percent of aberrant cells increased from 1.50 in the vehicle controls to 4.25 in the 200 mg/kg group ($p = 0.01$). Although in the second trial only a mean of 6.0 % of metaphases exhibited aberrations with the DMBA group, this still represented a significant increase ($p = 0.0004$).

Bone-marrow and peripheral blood micronucleus test . Shelby *et al.* (1993) reported only on the bone-marrow part of the NTP-sponsored testing of 1,3-D. According to the information provided by the NTP Archives, the testing actually utilized two study designs, with two trials being conducted for each design and 5-7 male mice (B6C3F1) being used per dose group, including vehicle and positive controls. The results of the full testing are described below.

The first design involved intraperitoneal injections of corn-oil solutions containing 1,3-D on three consecutive days, with sacrifice 24 hours after the last injection. Each of the test animals in the first design were used to obtain both bone marrow smears and peripheral blood smears. For both trials, the doses were 0 (corn oil), 31, 62.5 and 125 mg/kg, with 7,12-dimethylbenz[a]anthracene (DMBA) at 12.5 mg/kg used as the positive control. In both trials, there were no increases in the incidence of micronucleated polychromatic erythrocytes (PCE's) isolated from bone marrow or peripheral blood. By comparison, DMBA (12.5 mg/kg) significantly ($p < 0.003$) increased the incidence of micronucleated PCE's relative to vehicle controls by factors of 2.6 and 1.7 for the first trial using smears made from bone-marrow and peripheral blood, respectively; and in the second trial, the incidences of micronucleated PCE's were increased by factors of 2.7 and 3.7 using smears made from bone-marrow and peripheral blood, respectively.

For the second design, animals received a single intraperitoneal injection of corn-oil solutions containing 1,3-D. Also, the highest dose was greater, sacrifices in each trial occurred at 24 or 48 hours after the injection, and smears only were made from bone marrow. For the first trial with sacrifice at 24 hours after dosing, the doses were 0 (corn oil), 50, 100 and 200 mg/kg, with DMBA given at 50 mg/kg. For the first trial with sacrifice at 48 hours after dosing, the dose groups were the same except that there was no 50 mg/kg 1,3-D group. For the second trial with sacrifice at 24 hours after dosing, the doses were 0 (corn oil), 100, 150 and 250 mg/kg, with DMBA still given at 50 mg/kg. For the second trial with sacrifice at 48 hours after dosing, the dose groups were the same except that there was no 100 mg/kg 1,3-D group.

In both trials sacrificing animals 24 hours after one dosing, no significant increases in the incidence of micronucleated PCE's isolated from bone marrow were caused by 1,3-D. By comparison, DMBA (50 mg/kg) increased the incidence of micronucleated PCE's relative to vehicle controls by factors of 10.6 and 12.6 in the first and second trials, respectively.

However, with animals sacrificed at 48 hours after a single dosing, the incidences of micronucleated PCE's were increased by factors of 2.3 ($p \leq 0.01$) and 3.6 ($p \leq 0.001$) at the highest dose tested in the first and second trials, 200 and 250 mg/kg, respectively. Also, DMBA (50 mg/kg) increased the incidence of micronucleated PCE's relative to vehicle controls by factors of 14.0 and 9.3 in the first and second trials, respectively. As concluded by Shelby *et al*. (1993), the induction of micronucleated PCE's by 1,3-D appears to be dependent on test conditions, especially the following: doses of at least 150 mg/kg; the use of single dosing; and harvesting bone marrow at 48 hours after dosing.

III. *In vivo* testing in rats reported by Ghia *et al*. (1993), Kitchin *et al*. (1993) and Kitchin and Brown (1994)

A. Analysis of Ghia *et al*. (1993)

Preface. This study reported negative and positive findings for genotoxicity using three different *in vivo* assays. These assays will be discussed individually after introducing the following two important concepts regarding the test animals.

First, some of the testing in Ghia *et al*. (1993) involved rats that were not physiologically normal given that two-thirds of their liver had been removed 20 hours *before* their being orally dosed with 1,3-D. The partial hepatectomy was done to stimulate cell division, in the hope of facilitating micronuclei formation in hepatocytes of animals exposed to chromosome-breaking agents. However, such major liver reduction also initiates a slew of metabolic changes, including some that may affect the activation of promutagens and proclastogens. For example, Solangi *et al*. (1988) observed the following in rats in the first few days following a partial hepatectomy: an increase in hepatic heme oxygenase with a decrease in hepatic 5-aminolevulinic acid synthase, enzymes that control heme catabolism and synthesis, respectively; and decreases in hepatic cytochrome P450 content, accompanied by decreases in associated hepatic enzyme activities like aryl hydrocarbon hydroxylase, 7-ethoxycoumarin-O-deethylase and benzphetamine N-demethylase.

Second, gavage and i.p. injection were used as routes of exposure in Ghia *et al*. (1993). For both routes, the vehicle for 1,3-D was dimethyl sulfoxide (DMSO). Animals were dosed with DMSO solutions at 10 mL/kg; this constituted a DMSO dose of 11 grams/kg (140 mmole/kg).[25] It is reasonable to assume that significant amounts of DMSO will travel to the liver with both routes of exposure. For example, Kim *et al*. (2007) injected mice i.p. with DMSO (2.5 mL/kg) and observed that DMSO concentrations in the liver peaked at 26.7 µmole/g liver at 2 hr after injection, then declined with a half-life of approximately 2 hr.

The problem is that DMSO is not metabolically inert, as also demonstrated in Kim *et al*. (2007). Mice injected i.p. with just DMSO (2.5 mL/kg) then sacrificed 6 hr later exhibited a 34% in-

[25] The density of DMSO is 1.1 g/cm^3 and its molecular weight is 78.13.

crease in 4-nitrophenol hydroxylase activity ($p<0.001$) and a 19% increase in 4-nitroanisole O-demethylase activity ($p<0.05$). Kim *et al*. (2007) also studied the effects of coexposure to DMSO on the toxicities of dichloromethane (CH_2Cl_2) and carbon tetrachloride (CCl_4). Mice were injected i.p. with DMSO (1-5 mL/kg [14-70 mmole/kg]), followed 15 min later by an i.p. injection of either CH_2Cl_2 (6 mmole/kg) or CCl_4 (0.1 or 0.2 mmole/kg), using corn oil as the vehicle. The endpoint for CH_2Cl_2 was carboxyhemoglobin (COHb) in blood; this presumably resulted from CYP2E1-mediated hydroxylation of CH_2Cl_2, followed by spontaneous dehydrochlorinations that gave formyl chloride (Cl-CHO) and then carbon monoxide (CO), which reacted with hemoglobin. The peak level of COHb, which occurred 30-60 min after the injection of CH_2Cl_2, as well as the area under the curve were decreased in a dose-dependent manner by coexposure to DMSO. Using hepatic microsomes isolated from untreated mice, the microsomal metabolism of CH_2Cl_2 was shown to be inhibited by DMSO in a dose-dependent manner; Lineweaver–Burk plot of these data identified this as competitive inhibition. In contrast, the endpoint for CCl_4 was hepatotoxicity, as evidenced by increased liver enzyme activities[26] in serum from mice sacrificed 24 hr after injection of CCl_4. DMSO given at $\geq$ 2.5 mL/kg significantly ($p<0.05$) reduced each of the three serum activities that had increased as a result of the liver damage caused by the low dose (0.1 mmole/kg) of CCl_4. However, DMSO given up to 5 mL/kg did not attenuate any of the three serum activities that were each increased by the high dose (0.2 mmole/kg) of CCl_4. Using hepatic microsomes isolated from untreated mice, DMSO was shown not to inhibit the microsomal metabolism of CCl_4. Therefore, Kim *et al*. (2007) concluded that the DMSO inhibition of CCl_4-induced hepatotoxicity does not involve DMSO inhibition of the metabolic activation of CCl_4. The authors speculated that the mechanism possibly could involve inhibition of endogenous, signaling compounds like the eicosanoids.[27]

In vivo micronucleus assay on freshly isolated hepatocytes and polychromatic erythrocytes (PCE's) from bone-marrow and spleen using rats dosed after partial hepatectomy. These experiments were modeled after ones reported by Tates *et al*. (1980). Male Sprague-Dawley rats, 3-5 per dosing group (described below), underwent a two-thirds hepatectomy. Twenty hours later, animals were dosed in one of the following ways: gavaged once with 1,3-D at 125 mg/kg using DMSO as the vehicle (10 mL/kg); gavaged once with DMSO alone at 10 mL/kg (negative control); or injected i.p. once with N-nitrosodimethylamine (NDMA) at 10 mg/kg, using a vehicle that was not stated [28] (positive control). Animals were sacrificed 48 hr after dosing. Hepatocyte suspensions prepared after *in situ* collagenase perfusion of liver were dripped on slides,

[26] Three enzymes were assayed: aspartate aminotransferase, alanine aminotransferase and sorbitol dehydrogenase.

[27] Eicosanoids are endogenous compounds derived from arachidonic acid, e.g., prostaglandins, thromboxanes, leukotrienes.

[28] Examination of contemporary publications from the same research group as Ghia *et al.* (1993) indicates that the vehicle for i.p injection of NDMA at 10 mg/kg also was not stated (Martelli *et al.*, 1991; Allavena *et al.*, 1992; Mereto *et al.*, 1994), with one exception wherein the vehicle was identified as saline (Martelli *et al.*, 1996). Also, in Tates *et al*. (1980), the vehicle for i.p. injection of NDMA was not stated, but in

fixed and stained. For bone-marrow PCE's, one femur was flushed with fetal bovine serum and diluted cell suspensions thereof were cytocentrifuged onto slides, which were dried and stained. The method for isolating PCE's from spleen was not indicated; presumably, it also involved cytocentrifugation of cell suspensions onto slides.

With 1,3-D dosed at 125 mg/kg, there was no statistically significant increase ($p>0.05$) relative to the vehicle control group in the frequencies of micronucleated hepatocytes or micronucleated PCE's isolated from bone marrow or spleen. Also, with the 1,3-D treated group when compared to the vehicle control group, there was no change in the frequency of binucleated hepatocytes, the mitotic index of the hepatocytes, or the percentages of erythrocytes from bone marrow or spleen that were PCE's, as opposed to normochromatic erythrocytes. Such results would suggest that a higher dose of 1,3-D could have been tested. However, when 250 mg/kg was tried in this assay, several of the animals died within 24 hr of dosing.

Regarding the NDMA group (positive control), it served its minimal function, that of showing that a positive effect could be detected by this assay if it were present. Thus, with the NDMA group (i.p. injection) when compared to the DMSO-only group (gavage), the frequencies of micronucleated hepatocytes and micronucleated PCE's in bone marrow and spleen were increased by factors of 7.2, 14.4 and 10.1, respectively ($p<0.05$ for each). However, because the route of exposure was i.p. injection and the vehicle for NDMA was not stated to be DMSO (and may have been saline [see footnote 30]), the results with NDMA reported in Ghia *et al*. (1993) do not provide information about whether the oral exposure to DMSO at 10 mL/kg could have inhibited the metabolic activation of NDMA (and by extension, the activation of 1,3-D) in rats which were already compromised significantly due to having undergone a partial hepatectomy.

Unscheduled DNA synthesis (UDS) measured in hepatocytes isolated from treated animals. Male Sprague-Dawley rats, 3-4 per dosing group, were fasted overnight, then dosed in one of the following ways: gavaged once with 1,3-D at 125 mg/kg using DMSO as the vehicle (10 mL/kg); gavaged once with just DMSO at 10 mL/kg (negative control); or injected i.p. once with N-nitrosodimethylamine (NDMA) at 10 mg/kg, using a vehicle that was not stated [30] (positive control). Animals were sacrificed 3 hr after dosing and hepatocyte suspensions obtained after *in situ* collagenase perfusion of liver were used to prepare primary cultures. After cells attached,

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contemporary publications by one or more of the authors of Tates *et al*. (1980) the vehicle for i.p. injection of NDMA was saline (0.15 M NaCl) (Den Engelse *et al*, 1981) or phosphate-buffered saline (Tates *et al*., 1983). Therefore, it seems plausible that in Ghia *et al*. (1983), the vehicle for NDMA was not DMSO, the vehicle used for 1,3-D. If that is the case, the opportunity was missed to see if DMSO at 10 mL/kg would inhibit the induction of micronuclei by NDMA, e.g., by inhibiting its P450-mediated demethylation, an effect known for DMSO using hepatic microsomes from mice (Jeffery *et al*., 1988).

the medium was replaced with serum-free medium containing tritiated thymidine for incorporation into any newly synthesized DNA. After a 4 hr incubation, the radiolabel was washed out and medium now containing nonradiolabeled thymidine was added to the cultures for an overnight incubation. UDS was determined autoradiographically, with 100 hepatocytes being manually counted for each rat (50 cells per duplicate slides per rat). The endpoints for UDS were net nuclear grains (NNG, calculated as nuclear grains minus cytoplasmic grains) and percentage of cells showing DNA repair (defined as having an NNG of at least 5 grains).

No statistical analysis of the UDS data was provided in Ghia *et al*. (1993). However, the data analysis seems straightforward. With 1,3-D dosed at 125 mg/kg, there was no significant increase in UDS, both in terms of NNG (mean $\pm$ one standard deviation of 0.2 $\pm$ 2.0 and 0.5 $\pm$ 2.5 in the vehicle control group and 1,3-D group, respectively) and percentage of cells showing repair (1.7 $\pm$ 2.0 and 6.2 $\pm$ 6.6 in the vehicle control group and 1,3-D group, respectively).

Regarding the NDMA group (positive control), it served its minimal function, that of showing that a positive effect could be detected by this assay if it were present. With the NDMA group, NNG was 22.1 $\pm$ 12.4 and the percentage of cells showing repair was 92.8 $\pm$ 11.4. As discussed in the previous section, because NDMA was given by i.p. injection and the vehicle for NDMA was not stated to be DMSO (and may have been saline [see footnote 30]), the results with NDMA do not provide information about whether the oral exposure to DMSO at 10 mL/kg could have inhibited the metabolic activation of NDMA (and by extension, the activation of 1,3-D) in rats. Similarly, it is not clear from the NDMA testing that DMSO at such a large dose did not inhibit DNA repair, e.g., *via* inhibition of oxidative phosphorylation (hence ATP formation) in liver by the DMSO metabolite, dimethyl sulfide (Mhatre *et al.*, 1983).

Whether animals could have been tested at a higher dose of 1,3-D was not addressed by the authors. However, it seems reasonable that this could have been tried given that the animals were not hepatectomized, were sacrificed just 3 hours after dosing and were successfully gavaged at 250 mg/kg and sacrificed 24 hr afterwards in the alkaline-elution studies that are discussed next.

Alkaline-elution assay for detection of DNA breakage in cells isolated from various organs.

Male Sprague-Dawley rats, 3-12 per group, were fasted overnight, then dosed by gavage or i.p. injection with DMSO solutions of 1,3-D delivered using a dosing volume of 10 mL/kg. The 1,3-D dose levels were 0 (DMSO), 62.5, 125 and 250 mg /kg. In some experiments, animals were injected i.p. with one of three metabolism modulators (dissolved in corn oil or saline) before

being gavaged with 1,3-D: diethyl maleate (DEM), to reduce hepatic GSH content; buthionine-sulfoximine (BSO), to inhibit GSH synthesis; and 8-methoxypsoralen ("methoxsalen" [MS]), to inhibit cytochrome P450. Animals were sacrificed 1, 3 or 24 hr after 1,3-D dosing and liver, lungs, stomach, kidneys, brain and bone marrow were processed to obtain cell suspensions. DNA breakage[29] was assessed by placing cell suspensions on 5μm pore filters, lysing the cells with an anionic detergent which deposited double-stranded DNA on the filter, and then eluting the DNA through the filter using a strongly alkaline solution containing a chelating agent, which denatured the DNA into single strands. For this assay, the endpoint was the rate of DNA moving through the filter into the eluate. Suffice it to note that long, intact single strands elute more slowly than do broken single strands. The elution rate, K, was calculated using the equation: $K = (-\ln FR) / V$ where V is the eluate volume after pumping the eluting fluid at 0.13 mL/min for 100 min (hence V = 13 mL) and FR is the fraction of DNA remaining on the filter. As a first approximation, K is directly proportional to the frequency of DNA single-strand breaks. The data were analyzed statistically as the relative DNA elution rate, defined as the ratio of the treatment elution rate, K_t , to the negative-control elution rate, K_c, (i.e., K_t / K_c).

The main findings with this assay were the following. 1) In a dose dependent manner, each dose level of 1,3-D induced a statistically significant ($p < 0.02$) increase in DNA damage in liver and gastric mucosa, based on animals sacrificed 3 hr after being gavaged. [30] The maximal effects in liver and gastric mucosa were seen at 250 mg/kg: mean relative DNA elution rates were increased to 4.11 and 1.99, respectively. 2) In comparing sacrifice times, it appeared that the maximal amount of DNA damage was seen in liver and gastric mucosa when animals were sacrificed 3 hr after dosing, based on animals gavaged with 125 mg/kg. 3) In comparing routes of exposure, using animals dosed with 125 mg/kg and sacrificed 3 hr later, more DNA damage in liver was seen with oral exposure whereas more DNA damage in kidney was seen with i.p. injection (only organs with data allowing comparison). 4) No significant increase ($p > 0.10$) in DNA damage was seen in lung, bone marrow or brain. 5) 1,3-D at 125 mg/kg (gavage) reduced liver GSH content by 84% when assayed 1 hr after dosing (time of maximal effect) whereas when the animals were pretreated with DEM, liver GSH content 1 hr after the same 1,3-D dosing was reduced by 95%. However, when the endpoint was DNA damage in liver at 3 hr after 1,3-D at 125 mg/kg (gavage), pretreatment with DEM did not increase significantly mean relative DNA elution rate ($K_t / K_c = 3.22$), in comparison to the animals not exposed to DEM before being gavaged with 1,3-D ($K_t / K_c = 2.83$). Similarly, no increase in liver DNA damage was seen when animals were pretreated with BSO before being dosed with 1,3-D. MS was the only pretreatment to exert a

[29] The version of the assay used in Ghia *et al*. (1993) detects DNA strand breaks existing when cells were lysed on the filter and "alkaline labile" DNA sites. An example of the latter is when, due to high pH, DNA breaks at sites on the strand lacking a purine or pyrimidine base (abasic sites), e.g., from loss of guanine due to N7 alkylation. This version of the assay does not detect double-strand breakage or any crosslinking (DNA-protein or DNA-DNA).

[30] With kidney, lung, bone marrow and brain, data were provided only for animals dosed at 0 and 125 mg/kg, using i.p. injection and (or) gavage and sacrifice at 24 hr and (or) 3 hr after dosing.

significant effect on the liver DNA damage induced by 1,3-D (125 mg/kg by gavage): MS decreased mean relative DNA elution rate from 2.83 (no pretreatment) to 1.86 ($p = 0.01$). Ghia *et al*. (1993) concluded that in liver, cytochrome P450-mediated metabolism plays a significant role in the DNA damage caused by 1,3-D.

Assessment of Ghia *et al*. (1993). This study reported conflicting results regarding the genotoxicity of 1,3-D using assays designed to detect seemingly related activities--DNA breakage, chromosome breakage and DNA repair. There were dose responses using the alkaline-elution assay to detect DNA damage in liver and gastric mucosa as well as positive findings in kidneys at the one dose level studied. In liver, it was shown that pretreatment with an inhibitor of cytochrome P450 decreased the DNA damage induced by 1,3-D, thus indicating that some of the DNA damage depended on metabolic activation. By contrast, there were negative findings for the induction of micronuclei in liver cells and in erythroblasts in bone-marrow and spleen and negative findings for the induction of UDS in liver cells. However, there are good reasons to treat the negative findings as being provisional. First, all rats in this study were dosed with DMSO solutions of 1,3-D at 10 mL/kg. Although the negative controls received DMSO at 10 mL/kg by the same route used to deliver 1,3-D, this does not alter the fact that 1,3-D-treated animals, at each dose level, internalized much more DMSO than 1,3-D. To illustrate, at 125 mg/kg, the only dose tried in the negative testing, the dosing solution contained 123 moles of DMSO for every mole of 1,3-D. One problem with this is that DMSO is metabolized by cytochrome P450 and otherwise can inhibit cytochrome P450-mediated metabolism of other chemicals, possibly including 1,3-D. Second, in the testing involving the induction of micronuclei in liver cells and erythroblasts, the rats had livers that had been greatly reduced in size 20 hr before exposure to 1,3-D. As discussed already, in the time frame when the partially hepatectomized rats were dosed with 1,3-D, these rats likely had hepatic cytochrome P450 content and associated enzyme activities that were significantly reduced. Factoring in that these rats also were coexposed to a high dose of DMSO when dosed with 1,3-D, it would be surprising if the metabolism of 1,3-D was not significantly reduced, possibly so much so as to result in a negative test result. This point is valid notwithstanding the use of NDMA as a positive control because its route of exposure (i.p. injection) was different from the route used with 1,3-D (gavage) and the vehicle for NDMA was not stated to be DMSO (and may have been saline). Thus, the "positive" results seen with NDMA do not provide information about whether the oral exposure to DMSO at 10 mL/kg in the 1,3-D treated rats could have inhibited the metabolic activation of NDMA, and by extension, the activation of 1,3-D to clastogenic and (or) DNA damaging agents. As to why there were positive findings for DNA damage in liver using the alkaline-elution method when there were negative findings for UDS in liver--even though in both cases, rats had normal livers, were exposed to DMSO at 10 mL/kg, and were sacrificed 3 hr postdosing, the considerations include the following: different agents and (or) different mechanisms ultimately are responsible for the DNA damage detected by alkaline elution *versus* the DNA damage repaired during UDS;

1,3-D is “X-ray-like” in damaging DNA, inducing short-patch repair which is not detected well in the UDS assay; coexposure to the high DMSO dose inhibited DNA repair; testing at a higher level of 1,3-D or using different sacrifice times may have resulted in positive findings.

B. Analysis of Kitchin *et al*. (1993) and Kitchin and Brown (1994)

1,3-D was one of 111 chemicals that were tested by this research group at the USEPA Health Effects Research Laboratory at Research Triangle Park, North Carolina in the mid 1980’s to the early 1990’s (Kitchin *et al.*, 1992). The genotoxic endpoint was DNA damage in liver as measured using the alkaline-elution procedure. In the studies concerning 1,3-D, female Sprague-Dawley rats were dosed by gavage using corn-oil solutions. Animals were dosed twice, at 21 hr and 4 hr before sacrifice.[31] The dose levels at each of the two dosing times were 0 (corn oil), 0.094, 0.94, 9.4, 94 and 282 mg/kg; therefore, the total doses received in the 21 hr period before sacrifice were 0, 0.188, 1.88, 18.8, 188 and 564 mg/kg.[32] The testing consisted of four separate experiments, each involving one or several dose levels of 1,3-D as well as a corn-oil-only group; in some experiments, other test chemicals (also using corn oil as their vehicle) were tested, in addition to 1,3-D.

Liver was processed using ice-cold temperatures. After washing, tissue was minced, then lightly homogenized in a buffered, salty solution (1.5 grams in 6 mL). The homogenate was allowed to settle for 10 min, after which an aliquot of the homogenate (presumably dominated by intact hepatocytes, fragmented hepatocytes and their nuclei) was placed on filters for lysing. The alkaline-elution procedure that followed was similar to the method described previously in discussing Ghia *et al*. (1993), except for the following: the lysis solution was allowed to cover the filter for 48 hr before being drained; and the elution of DNA from the filter lasted for 14 hr. The former was done in the hopes of increasing the sensitivity of the assay for detecting DNA breakage by altering the flexibility and packing of the DNA (Nicolini *et al.*, 1985). The endpoint for assessing DNA damage was the fraction of DNA applied to the filter that was eluted. With livers from rats gavaged with corn oil only, the mean fraction of the DNA that eluted ranged from 0.103 to

[31] These dosing times were selected based on the hepatic endpoints measured in the study. DNA damage and changes in ornithine decarboxylase activity (in supernatant after high-speed centrifugation for liver microsomes) and glutathione content were expected to need 4 hr to manifest themselves whereas changes in cytochrome P450 and serum alanine aminotransferase activity were expected to need 21 hr to manifest themselves (Kitchin *et al.*, 1991).

[32] The doses tested and other information about the testing and results were confirmed by contacting Dr. Kirk Kitchin (USEPA).

0.213 (based on four groups, with 6-12 rats per group). In other words, 79-90% of the DNA from corn-oil-only animals typically remained on the filter at the end of the elution period.

The two highest doses, 564 and 188 mg/kg, caused significant increases in the mean fraction of DNA that was eluted: 0.414 and 0.268, respectively, *versus* 0.213 and 0.139 in their respective, corn-oil only group ($p < 0.01$ in both cases). Thus, both high doses increased the fraction of DNA that was eluted by a factor of about 1.9, in comparison to the values measured in their respective, concurrently-tested, corn-oil-only group. The middle dose, 18.8 mg/kg, was tested twice and although the fraction of DNA that was eluted was increased by 55% in one experiment and 26% in the other (relative to what was measured in the respective, corn-oil-only group), neither increase was significant ($p > 0.05$). A similar situation occurred with testing 1.88 mg/kg: two experiments were done, resulting in increases of 58% and 38%, neither of which was significant ($p>0.05$). At the lowest dose, 0.188 mg/kg, the mean fraction of DNA eluted was increased by 24% relative to the concurrently-tested, corn-oil-only group and that also was not significant ($p>0.05$).

As mentioned in footnote 15, four nongenotoxic, hepatic endpoints also were measured. However, these four endpoints were not measured in animals treated at the lowest dose, 0.188 mg/kg. One endpoint was the activity of ornithine decarboxylase (ODC), which converts ornithine to putrescine. ODC, the rate-limiting enzyme in the polyamine biosynthetic pathway, is under multilevel regulation, which, in turn, enables it to be induced rapidly by various stimuli for cell physiology, growth, and differentiation. The two highest doses, 564 and 188 mg/kg, increased mean ODC activity by factors of 76.8 and 5.3, respectively ($p<0.01$ in both cases) while the next two lower doses, 18.8 and 1.88 mg/kg, did not significantly increase ODC activity ($p>0.05$ in each case).

A second endpoint was a blood-derived one used to detect hepatotoxicity, increased serum alanine aminotransferase (ALT) activity. Some degree of hepatotoxicity appeared to have occurred in animals dosed at 564 mg/kg: their mean serum ALT activity, 20.5 IU/L, was greater than the mean of 13.8 IU/L measured in the concurrently tested, corn-oil only group ($p <0.01$). However, no hepatotoxicity was indicated in the animals dosed at 188 mg/kg: their mean serum ALT activity, 10.6 IU/L, was comparable to what was measured in their concurrently tested, corn-oil only group, 10.2 IU/L. Similarly, 18.8 and 1.88 mg/kg did not significantly increase serum ALT activity ($p>0.05$ in each case).

Another endpoint was hepatic cytochrome P450 content. This was measured only in the two highest dose groups. In animals dosed at 564 mg/kg, P450 content was decreased by 43% (not statistically significant) while in animals dosed at 188 mg/kg, no decrease was observed. However, other chemicals tested by this same research group, using the same experimental design, also induced a comparable decrease in hepatic P450 content and these decreases were considered to be significant, treatment effects. Carbon tetrachloride dosed at 30 mg/kg (total dose) resulted in a 39% decrease ($p<0.01$) (Kitchin and Brown, 1989); and iodoform dosed at 142 mg/kg (total dose) gave a 41% decrease ($p<0.01$) (Kitchin *et al.*, 1993). Therefore, it is possible that the decreased P450 content observed with 1,3-D at 564 mg/kg represents a marginal, treatment effect that was not found statistically significant as a result of the limited number of animals tested per group and the variability in the measurements.

The fourth endpoint was hepatic glutathione (GSH) content. At 564 and 188 mg/kg, GSH content was significantly decreased by 35% and 28%, respectively ($p<0.05$ in each case) whereas no decrease in GSH content occurred in the animals dosed at 18.8 or 1.88 mg/kg.

Assessment of Kitchin *et al*. (1993) and Kitchin and Brown (1994). The two highest dose levels of 1,3-D, 564 and 188 mg/kg, induced DNA damage to about the same degree in liver based on the alkaline-elution assay. The DNA damage at 564 mg/kg was associated with other hepatic effects, some of which may represent liver toxicity (large increase in ODC activity, slight increase in serum ALT, a nonsignificant decrease in cytochrome P450 content, a large decrease in GSH content). At 188 mg/kg, DNA damage of a similar degree still was observed; but this was accompanied now by lesser changes in the other hepatic endpoints (slight increase in ODC activity, no changes in serum ALT activity or P450 content, less of a decrease in GSH content). At 18.8 mg/kg, DNA damage possibly may have been induced but the increases in two experiments were not statistically significant; ODC activity, serum ALT activity and GSH content were not significantly affected at the middle dose. Looked at collectively, the data indicate that DNA damage was induced in liver by acute, gavage exposure to a non-hepatotoxic dose of 1,3-D (no increase in serum ALT activity, no decrease in P450 content). Also, review of the results for other chemicals (2-chloroethanol, iodoform, carbon tetrachloride, chloroform) tested in Kitchin *et al*. (1993) or in the references cited therein--using the same experimental design--confirms that chemicals that induce hepatotoxicity (increased serum ALT activity) do not necessarily induce DNA damage. Therefore, the DNA damage induced in liver by 1,3-DCP at 188 mg/kg appears to result from its being directly genotoxic and is not dependent of its producing first cytotoxicity in liver.

IV. Analysis of the *in vivo* genotoxicity testing of 1,3-D using transgenic (Big Blue) mice (Gollapudi and Cieszlak, 1997).

Since a synopsis of the testing was presented in the Summary of Toxicology for 1,3-D, it will not be repeated here. Suffice it to note that no mutagenic effect was observed in lungs or liver of the transgenic mice that had been exposed by inhalation to 150 ppm 1,3-D for a total of 10 exposure days, at 6 hr/day and 5 days/week, with sacrifice 17 days after the final exposure day. Rather, in this section, the reasons why the negative findings should be viewed as provisional will be summarized. The first part of this analysis relies largely on the OECD Guidelines for the Testing of Chemicals –Transgenic Rodent Somatic and Germ Cell Gene Mutation (OECD 488, dated July 26, 2013) and the references cited therein. Although the transgenic-animal testing described in record 162475 (Gollapudi and Cieszlak, 1997) was conducted in 1996 *versus* OECD 488 was promulgated many years later, the latter still constitutes a reasonable basis by which to evaluate the study. The second part of the analysis broaches the question of whether this assay can detect the various types of genetic damage that possibly could result from exposure to 1,3-D.

<u>Reasons (not in order of importance) why record 162475 should be considered insufficient for concluding that 1,3-D does not act as a mutagen in lung and liver in this transgenic-mouse gene mutation assay</u>.

1) In record 162475, the administration period (total time during which animals were exposed to test agent agent) was only 10 days. However, OECD 488 recommends 28 daily exposures, which under some circumstances (e.g., to increase assay sensitivity) may need to be expanded to 8 weeks or more.

2) OECD 488 recommends that the top dose should be the maximum tolerated dose (MTD), defined as a dose producing signs of toxicity such that higher dose levels would be expected to produce lethality. The selection of 150 ppm as the top dose in record 162475 was not specifically discussed, except to note that the highest dose tested in the inhalation cancer bioassay (Lomax *et al*., 1988), a level at which lung adenomas were significantly increased in male mice, was 60 ppm. In record 162475, organ weights were not measured and regarding body weight, no effect was observed during the two weeks involving exposure or during the postexposure period. The only treatment effect was the cageside observation of decreased activity, on study days 3-5 (1^{st} exposure week) and study days 8-12 (2^{nd} exposure week); decreased activity was not noted during the postexposure period.

3) In record 162475, the time between the end of exposure and the sacrifice of the animals, was 17 days. This is called the "expression time:" it is the time during which the unrepaired DNA lesions are fixed into stable mutations. However, OECD 488 appears to recommend a significantly longer expression time when the target cell populations in the organs being analyzed exhibit a slow turnover time; liver was given as one example thereof. In the case of organs with slow cell turnover, OECD 488 recommends a 31 day arrangement: 28 daily dosing days (during which fixation of mutations also will occur) followed by sacrifice three days later. However, OECD 488 also acknowledges that the maximum mutant frequency may not manifest itself in slowly proliferating tissues under this compromise protocol and, therefore, OECD 488 suggests the use of alternative protocols, e.g., 28 days of daily dosing followed by 28 days of expression time before sacrificing. Although OECD 488 did not mention it, lung also can be considered a slowly proliferating tissue. In lung, cell turnover is related to the need for replacement of aging or injured cells. Thus, the cell turnover is faster with the epithelial cells of the major airways than with the epithelial cells of the alveoli. For mice and rats, normal cell turnover time is 2-10 days for tracheal, bronchial and bronchiolar epithelium but for alveolar epithelium, it is 28-35 days (Bowden, 1983).

4) In record 162475, a full, positive control was not included in the testing. Instead the following "partial," positive control was employed: DNA isolated from liver and lungs of transgenic animals that had been treated in the past with the mutagen diethylnitrosamine (DEN) then stored at -80ºC at SRI International (Menlo Park, CA) was processed along with the DNA isolated in the Registrant's 1,3-D study. Presumably, the dosing of transgenic animals with DEN and the isolation of DNA were conducted at SRI International, although this matter was not specifically addressed. Rather, the only information provided was that male transgenic animals were gavaged daily for 5 days, at 15 mg DEN/kg body weight, and sacrificed 1 year later; why such a long expression time was used was not explained.

However, OECD 488 recommends the use of concurrently-conducted, positive-control animals. Although OECD 488 does allow for the "partial," positive control used in record 162475, it is contingent on the conducting laboratory's providing contemporary evidence demonstrating the following: competency in all steps of the assay; routine use of the assay; and ability to reproduce expected results from published data. The conducting laboratory was not one of the Registrant's laboratories; rather, the tissues were shipped to SRI International and the DNA was isolated and processed there. However, no contemporary evidence was provided in record 162475 concerning the conducting laboratory. Rather, it was noted that the person in charge of the work performed at SRI International was Dr. Richard Winegar, who, at the time of the 1,3-D study, was a contributor to the contemporary literature on mutagenicity testing with transgenic animals.

It can be noted that OECD 488 also allows (without offering justification) that when concurrently conducted positive controls are used, it is not necessary to administer the positive control by the same route as used with the test chemical. Therefore, regarding record 162475, OECD 488 would not require that the positive control be a chemical (or chemicals) that upon inhalation exposure induced mutations in lung and liver of the transgenic animals. The fact that if this were required by OECD 488, finding chemicals to serve as full, positive controls for both lung and liver as target organs when inhalation is the route of exposure would be a challenge should be underscored. One reason why this may be so with lung as the target organ is discussed next.

5) Not addressed in OECD 488 is the following issue which possibly impacts the detection of mutations in some organs. Several of the organs typically targeted in transgenic-animal testing are dominated by particular cell types (hepatocytes in liver; lymphoctyes in spleen; myelopoietic cells, erythropoietic cells and lymphocytes in bone marrow) or in the case of testes, the cells of interest, the germ cells, are extruded from seminiferous tubules. However, neither of these is the situation encountered with lung. Haies et al. (1981) morphometrically analyzed lung tissues from female Sprague –Dawley rats. Although a similar study using mouse lung tissues could not be found, this rat study suffices to illustrate the issue at hand. Restricting the analysis to alveolar septal lung tissue and excluding capillaries therein, the following cell types were found: type I cells, type II cells, endothelial cells, interstitial cells, and macrophages. Relative to the total number of cells counted, their respective percentages were: 7.5%, 14.5%, 43.0%, 31.8% and 3.2%. Therefore, in rats and presumably in mice as well, the cell types of the respiratory epithelium, which are the ones of interest for a lung carcinogen like 1,3-D, are not the dominant cell type contributing to the pool of DNA that is retrieved, packaged and analyzed for gene mutations. Rather, 78% of the cells are interstitial cells (largely fibrocytes), endothelial cells or macrophages. Furthermore, since type I cells do not divide (instead they replenish their numbers from type II cells that divide then differentiate), the actual target cells for this transgenic-animal, gene-mutation assay, the type II cells, constitute just 14.5% of the cells whose DNA is being processed. In record 162475, how exactly lung from the exposed animals was processed was not explained. Therefore, it is possible that a further dilution of the DNA pool occurred due to the addition of DNA from cells from the larger airways and associated connective tissue.

Searching the open literature for data or discussion of the lung issue described above did not identify any pertinent publications. However, that searching did indicate that only a few chemicals that induce lung cancer in an animal bioassay using inhalation exposure and that were tested using transgenic-animal gene-mutation assays following inhalation exposure were found to be mutagenic in lung. Therefore, presently only a few chemicals could serve possibly as a full,

positive control for studies of lung mutagenesis using inhalation as the exposure route. In the case of ethylene oxide (Sisk *et al.*, 1997), the experimental design showing significant lung mutagenesis in male B6C3F1 Big Blue mice involved 4 weeks of inhalation exposure to 200 ppm, 6 h/day, 5 days/week, with sacrifice at 8 weeks after the last exposure day. The increase in mutant frequency was marginal: from mean of 6.2×10^{-5} in the 0 ppm group to 9.1×10^{-5} in the 200 ppm group ($p = 0.046$). With that same experimental design, however, no mutagenesis was induced in bone marrow, spleen or germ cells by exposure to 200 ppm ethylene oxide.

Reasons why record 162475, even if taken as a *bona fide*, negative study, may still be considered insufficient for concluding that 1,3-D could not possibly be acting as a genotoxicant in lung and (or) liver

1) It was noted in OECD 488 that there are constraints on the ability of transgenic-animal gene mutation assays to detect certain deletions. Present transgenes respond to mutagens that induce base-pair substitutions, frameshift mutations and small deletions and insertions. However, large deletions (e.g., those larger than the *lacI* transgene carried in the Big Blue mice used in record 162475) are not readily detected as mutations in the transgenic-animal gene-mutation assays (Heddle *et al.*, 2000). An example of an agent inducing large deletions would be ionizing radiation (Hoyes *et al.*, 1998).

2) Transgenic-animal gene-mutation assays may not readily detect chemicals that act mainly by inducing chromosome aberrations and act only weakly at inducing point mutations. The same would apply to chemicals that are inactive themselves but are converted by metabolism to chromosome-breaking derivatives. Examples of direct-acting chemicals are the methylating agent, methyl methanesulfonate (Tao *et al.*, 1993) and the DNA crosslinking agent, mitomycin C (Wahnschaffe *et al.*, 2005).

APPENDIX VII. SUPPLEMENTAL DISCUSSION OF THE REACTIVITY AND GENOTOXICITY OF 1,3-D METABOLITES AND DEGRADATES

The metabolism of 1,3-D is summarized schematically in Figure III.1. 3-Chloroallyl alcohol, 3-chloroacrolein and 3-chloropropenoic acid (3-chloroacrylic acid) also are degradates appearing in soil after application of 1,3-D containing products (Belser and Castro, 1971).

3-Chloroallyl alcohol (3-CAA) $Cl\text{-}CH{=}CH\text{-}CH_2\text{-}OH$

Both isomers of 1,3-D spontaneously hydrolyze in water, producing the corresponding 3-CAA's (McCall, 1987). In sterile buffered water (pH 5, 7 and 9), the rate of hydrolysis was affected by temperature but not pH. Although the half life of 1,3-D in water at 20ºC was 11.3 days, the Arrhenius plot and associated equation indicated that the half life at 37ºC would be 1.2 days. Therefore, nonenzymatic hydrolysis is expected to produce 3-CAA slowly *in vivo* after 1,3-D has been internalized following an exposure.

The metabolic fate of 3-CAA *in vivo* or *in vitro* with metabolic-activation systems has not been characterized. Based on analogy to what is known or suspected for ally alcohol, it is plausible that 3-CAA is metabolized to its corresponding aldehyde, 3-CA , by alcohol dehydrogenases (Serafini-Cessi, 1972), catalase (DeMaster *et al.*, 1994) and possibly cytochrome P450 (Morgan *et al.*, 1982) and that 3-CAA is epoxidated by cytochrome P450 (Patel *et al.*, 1980), with the expected product being 3-chloroglycidol (1-hydroxy-3-chloro-2,3-epoxypropane).

Indirect evidence that epoxidation may be significant is suggested by the results from the *Salmonella* testing of 3-CAA. Connors *et al*. (1990) using the plate incorporation assay reported that in the absence of a metabolic activation system, 3-CAA was not mutagenic to TA100 but in the presence of S9, 3-CAA induced 46,630 revertants/µmole.[33] However, since 3-CA is not mutagenic towards TA100 (Basu and Marnett, 1984), the activation in Connors *et al*. (Connors *et al.*, 1990) cannot be due to metabolism to 3-CA. Therefore, this leaves epoxidation as a possible explanation for the S9-mediated activation of 3-CAA to a strong mutagen for TA100. This is assuming that 3-chloroglycidol behaves like structurally related epoxides in terms of being

[33] Based on the observation in Connors *et al*. (1990) of 5.04×10^5 revertants /mg when testing the *cis* isomer, and assuming 92.52 as the molecular weight for 3-CAA.

mutagenic to TA100, e.g., glycidol, epichlorohydrin (Sinsheimer *et al.*, 1993) and the epoxide of 1,3-D (Watson *et al.*, 1987; Schneider *et al.*, 1998a; Schneider *et al.*, 1998b).

3-Chloroacrolein (3-CA) Cl-CH=CH-CHO

As discussed above, 3-CAA produced from hydrolysis of 1,3-D could be metabolized several ways to 3-CA. Also, a metabolic pathway from 1,3-D to 3-CA seems plausible based on studies of cytochrome P450-mediated hydroxylation of chlorine-bearing, methylene carbons in ethyl chloride (Fedtke *et al.*, 1994) and 1,2-dichloroethane (McCall *et al.*, 1983). In both cases, the initially formed hydroxylated metabolite is presumed to dehydrochlorinate; consequently, what formerly was the chlorine-bearing methylene carbon is converted to an aldehyde. Thus, just as acetaldehyde and chloroacetaldehyde were formed from ethyl chloride and 1,2-dichloroethane, respectively, cytochrome P450-mediated hydroxylation of the methylene carbon of 1,3-D is expected to produce 3-CA.

Online search of the open literature identified only a few toxicological studies concerning 3-CA. Like acrolein, 3-CA is reactive at both of its ends, forming Schiff-base adducts at the carbonyl and Michael-addition adducts at carbon 3. However, like propynal[34] (and unlike acrolein), 3-CA can form two adducts on carbon 3 due to the presence of the chloro group on the double bond. Bartels *et al*. (2004) reported that a urinary metabolite isolated from animals exposed to 1,3-D or 3-CAA was 3,3-*bis*(S-[N-acetylcysteinyl])propan-1-ol. This "dimercapturate" presumably originated from the conjugation of two molecules of glutathione (GSH) with carbon 3 of 3-CA.[35] The same dimercapturate has been found in urine from animals dosed with 2-propyn-1-ol which is presumed to be metabolized to propynal (Dix *et al.*, 2001; Banijamali *et al.*, 2003).

3-CA is not mutagenic towards TA100 or TA98 but it does induce frameshift mutations in *Salmonella* tester strain hisD3052, inducing 890 revertants/µmole using the plate incorporation assay. This is different from what is observed with its unsubstituted analog, acrolein: it is mutagenic with TA100 but it is not mutagenic with hisD3052. The same differential mutagenicity seen with 3-CA is also seen with other acroleins substituted on carbon 3 with good leaving groups (methoxy, ethoxy, benzoyloxy) as well as with propynal. It has been speculated that this differential mutagenicity may be an indication that 3-substituted acroleins and propynal are

[34] HC≡C-CHO
[35] Carbon 3 of 3-CA originally was carbon 1 of 1,3-D.

crosslinking agents given the difference in excision-repair ability between hisD3052 (has *uvrB*) and TA100 (lacks *uvrB*) (Basu and Marnett, 1984).

3-CA as well as 3-benzoyloxyacrolein and propynal also give a positive response[36] in the "NBP test." These three are so reactive in this testing that the reaction can be run at room temperature (Basu and Marnett, 1984), opposed to the heat treatment used in the NBP test of 1,3-D. Consistent with its high reactivity as an alkylating agent in the NBP test, 3-CA as well as 3-benzoyloxyacrolein and propynal are very toxic to the bacterial tester strains. In general, it is expected that high reactivity of 3-CA means that it readily reacts with endogenous nucleophiles like thiols and amines in amino acids, glutathione, proteins and DNA.

Eder *et al*. (1990) have characterized some of the DNA adducts that are formed with acroleins. Acrolein and 2-haloacroleins form saturated, cyclic, 1,N^2-guanine adducts (called type I adducts) whereas 3-substituted acroleins form unsaturated versions of these adducts (called type II adducts). Unsaturation of the cyclic adduct, which means that there is conjugation present in the cyclic ring, imparts a characteristic change to the adduct: type II adducts possess strong fluorescence whereas type I are not fluorescent. The authors mentioned how they were able to use HPLC and a fluorescence detector to detect adduct formation in DNA after incubation of bacteria with 3-chloro-3-methylacrolein (3-chlorocrotonaldehyde). They also noted that the chemicals giving type I *versus* type II adducts show the differential mutagenicity discussed previously: type I adduct-forming chemicals are mutagenic towards TA100 but not towards hisD3052 whereas type II adduct-forming chemicals (including 3-CA) show the opposite—they are mutagenic towards hisD3052 but not towards TA100.

3-Chloropropenoic acid (3-CPA[37]) Cl-CH=CH-COOH

3-CA is expected to be metabolized by aldehyde dehydrogenase to 3-CPA, but presumably to a low degree given the high, innate reactivity of 3-CA with endogenous nucleophiles. Connors *et al*. (1990) tested the *cis*- and *trans*-isomers of 3-CPA using *Salmonella* tester strains TA 97 and TA102 in the plate incorporation assay. TA97 detects chemicals causing frameshift mutations while TA102 detects oxidative mutagens (e.g., x-rays, hydrogen peroxide and other hydroperoxides) and reactive aldehydes (e.g., formaldehyde, glyoxal, glutaraldehyde, and malondial-

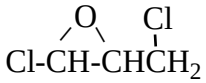
[36] An intense red color forms, absorbing at 487 nm. This is different from the purple color absorbing at 560 nm that is seen when 1,3-D is tested. Both color changes, however, indicate adduct formation with 4-(4-nitrobenzyl)pyridine.

[37] Named using the alternate name for acrylic acid, propenoic acid, to avoid confusion with abbreviations for 3-chloroally alcohol *versus* 3-chloroacrylic acid.

dehyde) (Levin *et al.*, 1982a; Levin *et al.*, 1982b). Both isomers of 3-CPA were stated to be mutagenic to both tester strains in the presence of S9. Test concentrations and resulting revertant counts, however, were not reported, except to note in general that the number of revertants per plate was increased by factors of 2-3 over the 282 revertants /plate observed with the solvent controls when testing concentrations in the nontoxic range, which was identified as <10 µg/plate (<94 nanomol/plate, assuming a molecular weight of 106.51)

It can be noted that *cis*-3-CPA was reported by Union Carbide scientists to be a cotton defoliant and crop desiccant (Herrett and Kurtz, 1963). Since *trans*-3-CPA did not exhibit this property and esters of *cis*-3-CPA (which would be expected to show some alkylating ability) were also inactive, the researchers speculated that the stereospecificity means that *cis*-3-CPA acts by inhibiting some aspect of the biochemical mechanism regulating leaf abscission. Whether there could be a target for *cis*-3-CPA in mammalians presumably is unexplored.

1,3-Dichloro-1,2-epoxypropane (1,3-D epoxide)



1,3-D was metabolized to 1,3-D epoxide in liver of male Swiss Webster mice that had been injected i.p. one time with a DMSO solution of 1,3-D. Also, 1,3-D was epoxidated by microsomes isolated from uninduced mice. 1,3-D epoxide is a direct-acting mutagen to TA100, inducing 17,000-37,000 revertants/µmole (depending on the isomer tested), using the plate incorporation assay. Nonenzymatic (and presumably epoxide-hydrolase-mediated) hydrolysis of the epoxide group in 1,3-D epoxide yields 3-chloro-2-hydroxypropanal (i.e., 3-chlorolactaldehyde). This may be toxicologically significant for three reasons. First, mechanistic studies of adduct formation seem to indicate that 3-chloro-2-hydroxypropanal is the ultimate species that alkylates deoxyguanosine. Second, metabolism of the R-enantiomer of 3-chloro-2-hydroxypropanal could produce the known kidney toxicant, (R)-3-chlorolactic acid (Schneider *et al.*, 1998a; Schneider *et al.*, 1998b). Third, if 3-chloro-2-hydroxypropanal were oxidized metabolically at its middle carbon (i.e., alcohol converted to carbonyl), the result would be another reactive and genotoxic metabolite, chloromethylglyoxal (Cl-CH_2-C[O]-CHO).

S-(3-chloro-2-propenyl)-N-acetyl-L-cysteine (3CP-NAc-Cys)

3CP-NAc-Cys is a major mercapturic acid excreted in urine following exposure to 1,3-D. Its formation generally is considered to be part of the detoxification process. However, there is some evidence that this pathway possibly can lead also to toxic effects in the urinary tract. Using the renal proximal tubule cell line derived from pig, LLC-PK1, and rat renal proximal tubular cells isolated from male Fischer 344 rats, Park *et al*. (1992) showed the following: 1) both isomers of 3CP-NAc-Cys [38] caused cytotoxicity in the LLC-PK1 cells (as judged by increased release of lactate dehydrogenase activity into the cell medium) and in isolated rat tubular cells (as judged by trypan blue exclusion); 2) using the *cis* isomer, the cytotoxicity to LLC-PK1 cells increased with a dose response over the concentrations tested (0.1-5.0 mM) and the cytotoxicity to isolated tubular cells increased with exposure time (up to 2 hr) as well as with increasing concentrations; and 3) the cytotoxicity of both mercapturic-acid isomers was greatly reduced in both types of cells by coexposure to methimazole (0.5 mM), which was used as an alternate-substrate, competitive inhibitor of flavin-containing monooxygenase (FMO). The latter results were viewed by the researchers as indicating that the cytotoxicity of these supposed, detoxification products was being caused by their metabolic activation in the cells, for example, by FMO-mediated S-oxidation. The researchers showed that the S-oxides of both of isomers of 3CP-NAc-Cys could be generated by chemical reaction with hydrogen peroxide or by metabolism using purified hog liver FMO in the presence of a NADPH generating system. Although the resulting S-oxides were sufficiently stable in water to be extracted and analyzed by HPLC, they were prone to undergo an interchemical reaction involving the oxygen of the S-oxide attacking the chlorine-bearing carbon of the propenyl group in the same molecule, a reaction called a [2,3]-sigmatropic rearrangement. Importantly, the resulting product of the rearrangement also is unstable: it decomposes into cysteine and acrolein--a reactive, genotoxic aldehyde that has been shown to initiate bladder cancer in F344 rats (Cohen *et al.*, 1992).

[38] 3CP-NAc-Cys isomers are defined by the propenyl group attached to the thiol in cysteine.

APPENDIX VIII. DERIVATION OF THE 1,3-D CANCER POTENCY FACTORS USED TO CALCULATE AMBIENT LIFETIME CANCER RISK

(A) Portal-of-Entry Effect:

Incidence of bronchioalveolar adenomas in male mice exposed to 1,3-dichloropropene via inhalation

Administered dose (mg/m^3)	Purity and duration adjusted dose[a] (mg/m^3)	Human equivalent concentration (HEC)[b] (mg/m^3)	Tumor incidence
0	0	0	9/49
22.7	3.7	12.7	6/50
90.8	15	51.6	13/49
272	45	154.8	22/50

[a]Correction for purity of formulation concentration (92%) and correction for intermittent exposure to continuous exposure: 22.7 mg/m^3 × 0.92 × 6/24 hours × 5/7 days = 3.7 mg/m^3.
[b]Correction for thoracic effects using RGDR(TH) of 3.44 (male mouse).

Thoracic Region: $RGDR(TH) = (MV_a/S_a)/(MV_h/S_h)$

where RGDR(TH) = regional gas dose ratio for the thoracic area of the lung
MV_a = animal minute volume (mouse [male] = 0.044 L/min)
MV_h = human minute volume (13.8 L/min)
S_a = surface area of the thoracic region of the animal lung (mouse = 503.5 cm^2)
S_h = surface area of the thoracic region of the human lung (543,200 cm^2).

Using default values, RGDR(TH) = (0.044/503.5)/(13.8/543,200) = 3.44.

Using benchmark dose modeling, the BMC_{10} or $LEC_{10\ (HEC)}$ = 25.86 mg/m^3
where $LEC_{10(HEC)}$ is the 10% extra risk at human equivalent concentration of LEC

Air Unit Risk (AUR):

1. Using Benchmark Dose Modeling
   AUR (95% UCL on risk at 1 ppm) $= 0.1/LEC_{10(HEC)\ (unit:\ ppm)}$
   $= 0.1/(25.86\ [mg/m^3].24.45L/110.98g)$
   $= 0.1/5.7$ ppm
   $= 0.018\ (ppm)^{-1}$

2. Using Linearized Multistage Modeling
   AUR (95% UCL on risk at 1 $\mu g/m^3$) $= (CPF_H \times BR\ m^3/day)\ /(BWt\ [kg] \times CF)$
   where:
   CPF_H = Cancer Potency Factor of Humans
   CF = Conversion factor (1000 μg/mg)
   BR (m^3/day) = Breathing Rate
   BR/BWt = Normalized Breathing Rate (nBR) (0.28 m^3/kg-day)

(Andrews and Patterson, 2000)

Rearrange the equation:

CPF_H = (AUR x CF)/nBR
= (3.87 x 10^{-6} [μg/m^3]$^{-1}$ x 1000 [μg/mg])/0.28 (mg/kg/day)$^{-1}$
= 0.014 (mg/kg/day)$^{-1}$

(B) Systemic Effect:

Incidence of bronchioalveolar adenomas in male mice exposed to 1,3-dichloropropene via inhalation

Administered dose (mg/m^3)	**Purity and duration adjusted dose[a] (mg/m^3)**	**Human equivalent concentration (HEC)[b] (mg/m^3)**	**Tumor incidence**
0	0	0	9/49
22.7	3.7	3.7	6/50
90.8	15	15	13/49
272	45	45	22/50

[a]Correction for purity of formulation concentration (92%) and correction for intermittent exposure to continuous exposure: 22.7 mg/m^3 × 0.92 × 6/24 hours × 5/7 days = 3.7 mg/m^3.
[b]Correction for Systemic Effect RGDR of 1.

Using benchmark dose modeling, the BMC_{10} or $LEC_{10\ (HEC)}$ = 7.49 mg/m^3
where $LEC_{10(HEC)}$ is the 10% extra risk at human equivalent concentration of LEC

Air Unit Risk (AUR):

1. Using Benchmark Dose Modeling
   AUR (95% UCL on risk at 1 ppm) = 0.1/$LEC_{10(HEC)\ (unit:\ ppm)}$
   = 0.1/(7.49 [mg/m^3].24.45L/110.98g)
   = 0.1/5.7 ppm
   = 0.061 (ppm)$^{-1}$

2. Using Linearized Multistage Modeling
   AUR (95% UCL on risk at 1 μg/m^3) = (CPF_H x BR m^3/day) /(BWt [kg] x CF)
   where:
   CPF_H = Cancer Potency Factor of Humans
   CF = Conversion factor (1000 μg/mg)
   BR (m^3/day) = Breathing Rate
   BR/BWt = Normalized Breathing Rate (nBR) (0.28 m^3/kg-day)
   (Andrews and Patterson, 2000)

Rearrange the equation:

CPF_H = (AUR x CF)/nBR
= (13.36 x 10^{-6} [μg/m^3]$^{-1}$ x 1000 [μg/mg])/0.28 (mg/kg/day)$^{-1}$
= 0.048 (mg/kg/day)$^{-1}$

APPENDIX IX. CDPR evaluation of the air dispersion modeling tool, SOFEA 2

Department of Pesticide Regulation

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DATE: August 12, 2015

SUBJECT: EVALUATION OF THE AIR DISPERSION MODELING TOOL SOFEA2

Background:

The air dispersion modeling tool SOFEA2, developed by Dow AgroSciences LLC (Dow), has been the subject of CDPR review since February 2014. The focus of the SOFEA2 modeling tool is estimating air concentrations of 1,3-Dichloropropene (1,3-D) associated with applications of the fumigant. The SOFEA2 model has been revised several times since the earliest version (dated December 31, 2013) was submitted for review. New data volumes were submitted for review following each revision. Various errors in the SOFEA2 model have been corrected with each new version. Dow also conducted a 14.5 month air monitoring study to collect measured air concentrations with the objective to validate the SOFEA2 model (Rotondaro and van Wesenbeeck, 2012). EM staff performed a complete review of the SOFEA2 model and the DOW validation analysis in data volume 50046-0210 (ID263794) (Johnson, 2014). The following technical deficiencies were noted:

1) SOFEA2 under estimates higher air concentrations

The Johnson (2014) evaluation of the March 21, 2014 version of SOFEA2 was confined to air concentrations measured and modeled at the 9 air monitoring locations in the center of the 9 townships from the Rotondaro and van Wesenbeeck, (2012) study. Based upon that analysis, Johnson (2014) concluded that "…SOFEA2 does a relatively poor job of estimating concentrations in both time and space."

2) SOFEA2 incorporates a version of the CHAIN2D model called VEFE which was assumed to have been used to generate the air concentrations presented in vol 50046-0210.

This current evaluation reviews the most recent version of SOFEA2 that includes: 1) corrections and improvements related to the Johnson (2014) review, 2) a new approach to more fully account for the effect of meteorological variables on air concentrations, and 3) additions to SOFEA2 to account for the influence of applications made outside of the 9 township area monitored by Dow on the measured and modeled 1,3-D air concentrations.

Evaluation:

The SOFEA2 model will be used as a prospective modeling tool. Specifically, the SOFEA2 model will be used to generate annual average air concentrations associated with the use of 1,3-D over many years in a region. The minimum geographic area of interest is the Public Land Survey System "township" (survey township) which is a 6 mile by 6 mile square parcel of land in California. The simulated annual average 1,3-D air concentrations are then used in risk analysis. Thus, the evaluation of the SOFEA2 model is in the context of how well the SOFEA2 model can be argued to capture the potential high concentrations associated with the use of 1,3-D. Air concentrations at any one specific point within a township is not of interest because the exact location of air concentrations within the township is not important in the risk analysis.

The Dow monitoring study (Rotondaro and van Wesenbeeck, 2012) was conducted over a 3 x 3 township area. This is a geographic scale of 18 miles by 18 miles. Nine air samplers were employed within this monitoring domain - one air sampler in the center of each township. Considering the scale of the modeling domain, this is an extremely sparse sampling set with which to validate the SOFEA2 model using air concentrations matched in space and time. However, the nine air sampler results can be used to explore whether SOFEA2 is generating maximum air concentrations sufficiently high to be argued that the worst case air concentrations have been captured. Due to the sparse sampling, it can be assumed that the maximum air concentration during the monitoring study was not captured by the air samplers. Therefore, the maximum air concentration is unknown and likely larger than the maximum measured air concentration. Furthermore, the design of the Dow monitoring study necessitates keeping all 9 townships together as a single domain of interest. The questions then are: 1) Does the model produce air concentrations at least as high as those measured and 2) Does the model produce annual average 1,3-D air concentrations that reflect the actual distribution of annual 1,3-D air concentrations during the monitoring study. The answers to both of these questions are highly dependent on the density of the receptor grid used to run the SOFEA2 model. A 9 receptor modeling grid that only represents the locations of the 9 air monitored locations within the 9 township (18 mile by 18 mile) area is not sufficient to evaluate the SOFEA2 model performance in the context of a prospective risk assessment scenario. Modeling with such a sparse receptor grid is extremely unlikely to adequately characterize the actual distribution of air concentrations in 9 township model domain.

The Dow monitoring was conducted as continuous 72-hr sampling periods over 14.5 months. This model evaluation will examine the period average, which is a 14.5 month average for two reasons: 1) the SOFEA2 model would not run to completion (this will be discussed further below) and 2) the end use of the data is the risk assessment process using the annual averages. The 14.5 month averages can be used as a surrogate for the annual average in this initial model evaluation.

An independent SOFEA2 model run at CDPR was used to evaluate the model. Results presented in the data volume guided the CDPR evaluation but are not the focus of this evaluation. As stated earlier, the distribution of 1,3-D air concentrations generated by the SOFEA2 model is highly dependent upon the density of the receptor grid. For multi-year prospective model runs in a 3x3 township model domain DOW has used a grid of 10,000 receptors (100x100 receptor frame) with a resulting spacing between receptors of about 290m (the data volume on page 23 states 11,664 receptors but that receptor grid includes extra receptors along the edges due to a bug in the receptor generating algorithm. That bug will be fixed in the next version of SOFEA). Using a 10,000 receptor grid should continue to be the practice for prospective model runs. For the DOW validation run, reported in the data volume, a receptor grid of 2500 receptors (50x50 receptor frame) spaced approximately 580 m apart was used. Initial evaluations using contour plots in SURFER software indicate that the 580 m receptor spacing is not sufficient to characterize the highest concentrations produced by the SOFEA2 model. The focus of SOFEA model validation has been whether the SOFEA2 model is sufficiently capturing the highest air concentrations. The validation model runs are retrospective rather than prospective so a very dense prospective receptor grid is not initially required. A 10,000 receptor grid generates a very large output file and is not absolutely necessary if a less fine grid of receptors spaced a little farther apart shows the SOFEA2 model captures the magnitude of the air concentrations in the 9 township area. Thus, for the initial validation a CDPR SOFEA2 run was performed using 5660 receptors (75x75 receptor frame) spaced approximately 400 m apart.

The actual CDPR model run was conducted outside of the SOFEA2 GUI because the SOFEA2 model would not run the 5600 receptor validation run to completion. The post-processing portion of the SOFEA2 run did not complete, despite many attempts to locate and fix errors that might be causing the problem. There are some serious bugs in the SOFEA2 GUI that must be corrected before this model can be used routinely at CDPR. However, this validation of SOFEA2 is a “proof of concept” with the objective being: does the ISCST model, using the SOFEA2 generated input file and the Merced weather file with the mixing height algorithm, produce air concentrations that reflect the conditions observed in the 3x3 township area monitored by DOW for 14.5 months. The post processing done by SOFEA2 after the ISCST run is not needed to achieve this objective. SOFEA2 post processing includes reporting for the entire 14.5 month period the 1 hr air concentrations at each receptor, finding the 72 hour air concentrations for each receptor, and presenting various graphical analyses. All the post processing is conducted in Excel. The post processing results are not necessary to demonstrate the proof of concept

condition has been met. Thus, the procedure to obtain the output was as follows: 1) generate the 5660 receptor grid within SOFEA2, 2) run the SOFEA2 model in validation mode to obtain the ISCST input files needed for the run, 3) run the ISCST model in a separate folder using the input files generated by SOFEA2 with the mixing height corrected weather file, 4) analyze both the weather file and the ISCST period average output file.

Analysis of the mixing height corrected weather file revealed two significant errors in the DOW processing of the weather data: 1) the stability classes are not correctly assigned in some cases, and 2) the mixing heights are miss-assigned by the mixing height algorithm.

Briefly, stability classes and mixing height characterize the turbulence and degree of vertical mixing in the atmosphere. There are 6 Pasquill stability classes categorized in classes from 1 to 6. Stability class 1 is the most unstable (the most vertical mixing), while stability class 6 is the most stable (the least vertical mixing). The progression from stability class 1 through 6 depends on the angle of the sun (time of day), degrees of cloud cover, and the wind speed. Stability classes 1 – 4 occur during the day. Stability classes 4-6 occur at night. The Pasquill stability classes are used by the ISCST3 model. The mixing height is defined as the height above ground within which the atmosphere can mix vertically (Turner, 1994). The higher the mixing height, the greater the potential to disperse a pollutant, all other factors held constant.

Table 1 shows a summary of stability class by hour of the day. Stability classes 1(very unstable) and 2 (unstable) are characteristic of warm, sunny days late in the morning into early afternoon. The Solar elevation angle required for stability class 1 is an angle equal to or greater than 60 degrees above the horizon (Zanetti, 1990). Stability class 2 requires a solar elevation angle between 35 degrees and 60 degrees above the horizon. Even at the summer solstice of June 21 the solar angle at 1000 hrs is 62 degrees, just satisfying solar elevation conditions for stability class 1. Therefore, stability class 1 should not occur earlier than 1000 hrs. The same solar elevation requirements must be met in the late afternoon. In addition, the wind speed cannot be greater than 3 mph together with the solar elevation of 60 degrees or greater for a stability class 1 to be assigned to an hour. Yet, Table 1 shows many hours of stability class 1 both early in the morning and late in the afternoon. Stability class 2 also is assigned in hours where it clearly cannot occur on the environment. Stability classes 5 and 6 should only occur when the sun is below the horizon. Yet stability class 5 occurs in every hour and stability class 6 occurs in 23 of the 24 hours of the day. The mistakes in stability class assignment will tend to reduce the air concentrations estimated by SOFEA2 because there are too many hours with very unstable and unstable atmospheric conditions. The mistakes in assignment of stability classes 5 and 6 affect less hours than stability classes 1 and 2. Therefore, with respect to model validation this would tend to cause the match of measured to modeled showing that the SOFEA2 model underestimates the air concentrations. A quick check of the Merced weather file shows that stability classes change by more than one class per hour. This cannot be allowed (Johnson et al., 1999). The stability class algorithm must also be corrected so that stability classes do not change

by more than one stability class per hour. For example, just before dawn if an hour is assigned stability class 6, the next hour must be 5 and the hour after that stability class 4.

Table 1. Summary of stability class assignment by hour in the MERC2010_2012_MH.met file supplied by Dow AgroSciences.

Hour	Stability Class						Total
	1	2	3	4	5	6	
0100	0	0	0	51	65	630	746
0200	0	0	0	48	62	636	746
0300	0	0	0	41	84	621	746
0400	0	0	0	53	66	627	746
0500	0	0	0	47	70	629	746
0600	150	5	0	47	88	456	746
0700	150	121	22	81	145	227	746
0800	151	283	89	150	47	26	746
0900	154	332	170	63	23	4	746
1000	188	399	110	44	2	3	746
1100	311	295	111	25	2	2	746
1200	367	252	97	28	1	1	746
1300	390	221	102	31	1	1	746
1400	395	218	98	34	0	1	746
1500	356	239	111	38	2	0	746
1600	301	268	135	40	1	1	746
1700	226	310	164	42	4	0	746
1800	176	327	164	53	6	20	746
1900	155	142	111	109	31	198	746
2000	146	26	57	154	75	288	746
2100	0	0	19	151	123	453	746
2200	0	0	0	123	103	520	746
2300	0	0	0	72	121	553	746
2400	0	0	0	55	88	603	746
Total	3616	3438	1560	1580	1210	6500	17904

The Dow mixing height adjustment algorithm is invoked when wind speed is 1.0 m/s (or less but wind speeds less than 1.0 m/s are set to 1.0 m/s). However, the adjustment should also be dependent upon the solar angle and/or the solar radiation. The Dow mixing height adjustment algorithm clearly does not distinguish between night and day hours (Table 2). This leads to the lowest median mixing height occurring during calm wind conditions in daylight hours. This is not true in the environment (Schnelle and Dey, 2000). In fact, calm winds and daylight hours

combine to produce some of the highest mixing heights. Wind speeds of 1.0 m/s during the day assigns stability class 1 (very unstable). Very unstable atmospheric conditions have the highest mixing heights of the day.

Thus, the main issues with the Dow mixing height adjustment algorithm are:
1) Stability classes 1,2, and 3 should not have a mixing height adjustment at all.
2) The lowest mixing heights should happen predominantly at night or at the transition hours around sunset and/or sunrise.
3) Stability class 6 should have the lowest median mixing height.

Table 2. Summary of adjusted mixing height by hour in the MERC2010_2012_MH.met file supplied by Dow AgroSciences.

Hour	Stability Class						Total
	1	2	3	4	5	6	
0100	*	*	*	*	12.80	16.40	16.35
0200	*	*	*	*	24.20	17.30	17.40
0300	*	*	*	*	8.10	15.80	15.80
0400	*	*	*	*	23.00	16.65	16.95
0500	*	*	*	34.00	24.90	17.80	18.25
0600	31.10	19.60	*	20.30	30.10	10.20	18.10
0700	31.20	17.50	32.30	28.20	25.15	4.70	16.90
0800	31.25	8.90	28.20	24.40	7.65	1.95	16.35
0900	29.50	9.00	17.40	11.80	2.00	1.60	13.20
1000	29.05	7.40	15.15	1.95	*	*	11.80
1100	27.40	7.80	1.85	*	*	*	13.20
1200	18.80	4.10	*	*	*	*	13.40
1300	18.40	5.10	*	*	*	*	13.50
1400	17.40	4.00	*	*	*	*	9.60
1500	16.80	4.50	*	*	*	*	7.50
1600	19.20	5.60	*	*	*	*	7.40
1700	22.95	6.30	5.20	*	2.80	*	9.60
1800	19.90	5.70	7.15	*	*	2.10	6.90
1900	30.00	7.85	2.60	2.30	5.20	7.10	7.90
2000	31.90	31.30	11.75	5.70	2.05	7.30	10.20
2100	*	*	*	12.70	5.70	13.20	12.70
2200	*	*	*	31.30	12.50	13.45	13.60
2300	*	*	*	*	24.15	13.35	13.60
2400	*	*	*	23.70	2.00	14.15	13.90
Total	27.4	7.3	10.45	15.85	19.35	13.3	14.4

*No hours

The issues with stability class assignment and the mixing height adjustments prevent a firm conclusion with respect to the ISCST validation scenario modeling results. The stability class

errors would tend to decrease the modeled air concentrations. The mixing height adjustment errors would likely increase the modeled air concentrations during the day but decrease them at night, relative to if they mixing height were correctly assigned to the same adjusted hours. As a result of these errors only a preliminary assessment of the modeled validation scenario air concentrations can be given.

As stated above, the SOFEA2 model itself would not run to completion. The SOFEA2 model did run the ISCST model to completion for the validation run but would not successfully perform the post processing. As a result of the SOFEA2 model "bombing" during the post processing, the output files produced by the ISCST model run did not get closed and, thus, were unavailable for analysis. The SOFEA2 post processing, while interesting, is not needed for the initial validation. SOFEA2 did successfully generate the input files needed to run the ISCST model outside of SOFEA2. For the purposes of evaluating the annual (or 14.5 month period) averages it is sufficient to use the modeling results directly from the ISCST model. No additional post processing is required. Table 3 summarizes the measured and modeled 14.5 month averages for each of the 9 monitored locations. All 9 modeled period averages for the monitored locations were within a factor of 2 of the measured 14.5 month measured air concentrations. The simplest metric to evaluate an air dispersion model is to compare the ratio of measured to modeled air concentrations. For regulatory purposes, an air dispersion model is considered "acceptable" if modeled air concentrations are within a factor of 2 of the measured air concentrations (Pratt et al., 2004).

Table 3. Comparison of measured and modeled 1,3-D air concentrations (ug/m^3)

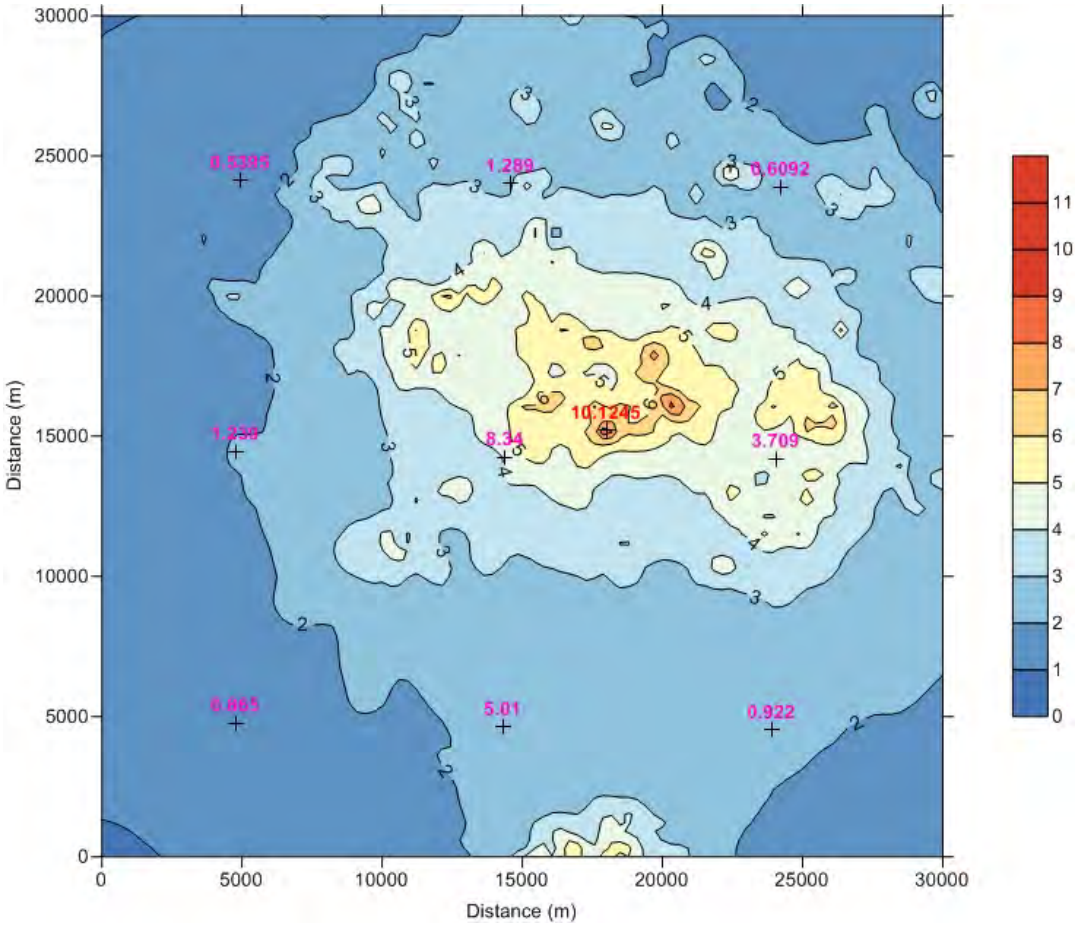
Township ID	14.5 month measured average air concentration (ug/m^3)	14.5 month modeled average air concentration (ug/m^3)	Measured/Modeled
1	0.8650	1.5278	0.57
2	5.0100	2.5171	1.99
3	0.9220	2.2478	0.41
4	1.2390	1.9790	0.63
5	8.3400	4.2815	1.95
6	3.7090	4.1197	0.90
7	0.5395	1.7330	0.31
8	1.2890	3.0171	0.43
9	0.6092	2.4413	0.25

The USEPA modeling guidelines acknowledges that air dispersion models are better at estimating longer term average air concentrations than short term (USEPA, 2005). In addition,

air dispersion models can be expected to reasonably match the magnitude of the maximum concentrations in a given area over a chosen period of time but cannot be expected to match exact locations (USEPA, 2005). This is due to uncertainties in model inputs. For example, errors in location of a plume due to meteorological data uncertainties and other data input errors can cause a 50% or more error in the estimation of an air concentration at a fixed location (Pasquill, 1974). Figure 1 shows the contour plot of ISCST modeled air concentrations for the validation scenario. It should be noted that these results are highly dependent upon the receptor grid density (as discussed above).

The results in Figure 1 are for the 5660 receptor grid. If the 10,000 receptor grid had been used it is likely even higher and more numerous maximum modeled air concentrations would have been found. The maximum measured air concentration of 8.34 ug/m^3 was exceeded by the modeled receptor concentration of 10.12 ug/m^3 at 2.3 miles from the measured location. A second model receptor showed a maximum modeled air concentration of 8.58 ug/m^3. The contour plot shows that there are several areas in the model domain where the modeled concentrations are in the 8 ug/m^3 range. Also shown is that some measured locations are underestimated and other are over estimated. But in the context of the regional concentrations the ISCST Merced validation scenario run captures the measured maximum air concentrations, just not in the exact locations where they were measured. The effect of the 1,3-D applications that were made just outside the 3 x 3 township area can be seen on the lower end of the plot. This demonstrates why it is important to include those applications both in the Merced validation scenario and prospective SOFEA2 runs.

Figure 1. Contour plot of ISCST modeled Merced validation scenario 14.5 month period average 1,3-D air concentrations (ug/m^3). Black crosses are the locations of the monitored air concentrations. Purple text are the measured 14.5 month 1,3-D air concentrations (ug/m^3). This axes show the 18 mile by 18 mile area as defined by the Dow study (in meters for ISCST modeling purposes), with the southwest corner as (0 m, 0 m). This coordinate system is not GIS based, instead it is referenced for the ISCST model with respect to the southwest corner of the model domain.



Comparing the distribution of measured 1,3-D concentrations to the modeled 1,3-D concentrations directly is problematic because only 9 locations were monitored. The monitoring grid was extremely sparse. Thus, the distribution of air concentrations fit to the measured values appears to over-estimate what the highest measured 1,3-D air concentrations would have been had a more comprehensive monitoring network been employed (Figure 2). A 99.99 percentile of 1,3-D air concentration of 60.3 ug/m^3 and 99.0 percentile of 15.5 ug/m^3 does not seem realistic. . It should not be assumed that concentrations in these ranges occur without measured concentrations in that range. The uncertainty in the measured air concentration probability distribution is evident from the width of the 95% confidence interval on the probability distribution. The lower 95% confidence interval values for this probability distribution are 9.3 ug/m^3 for the 99.99 percentile and 4.3 ug/m^3 for the 99.0 percentile. These air concentrations are in line with the modeled probability distribution (Figure 3). The 95% confidence intervals on the modeled concentration probability distribution are extremely narrow. This is because 5660 receptors comprise the input to fit that probability distribution. This is a very large input data set to characterize the distribution of air concentrations in the 9 township modeling domain so there is very little uncertainty in the shape of the distribution.

Figure 2. Probability plot of measured 1,3-D 14.5 month average air concentrations (ug/m^3).

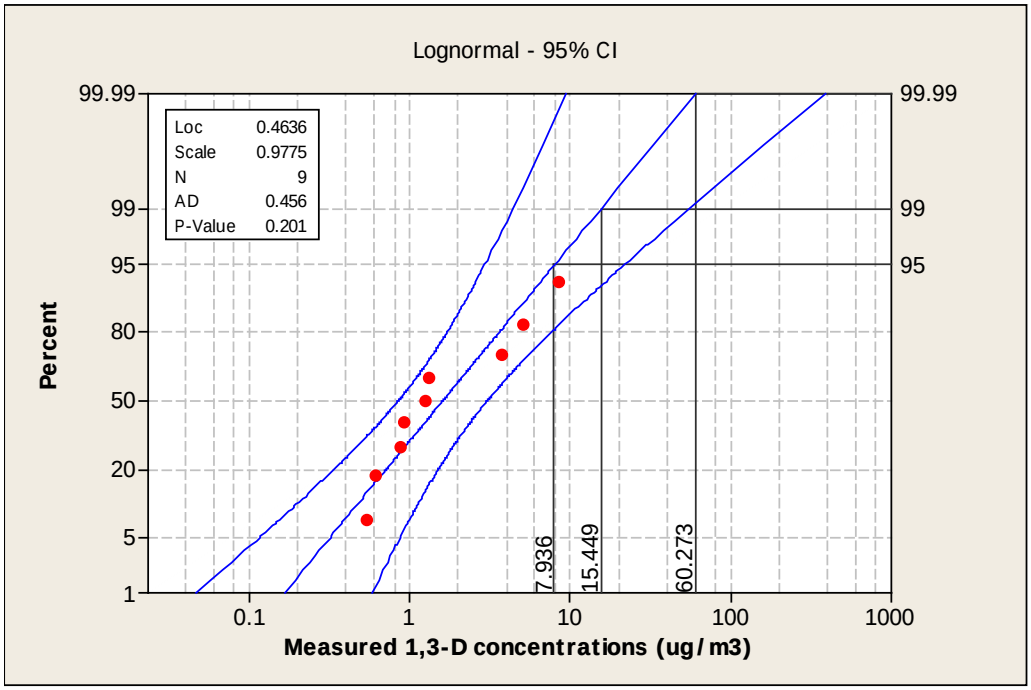


Figure 3. Probability plot of modeled 1,3-D 14.5 month average air concentrations(ug/m^3)..

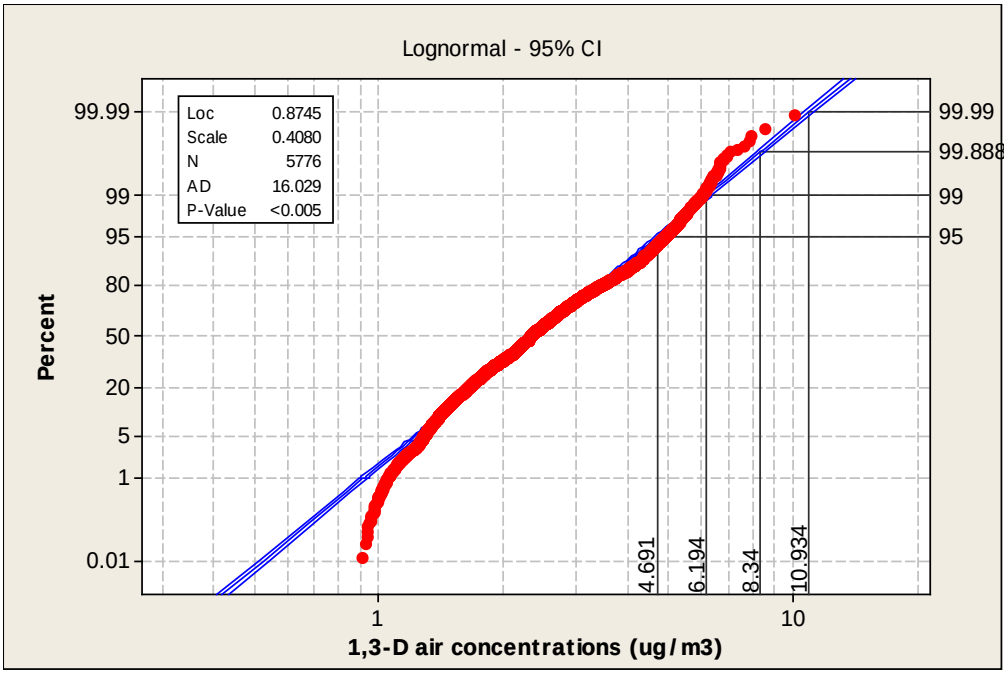
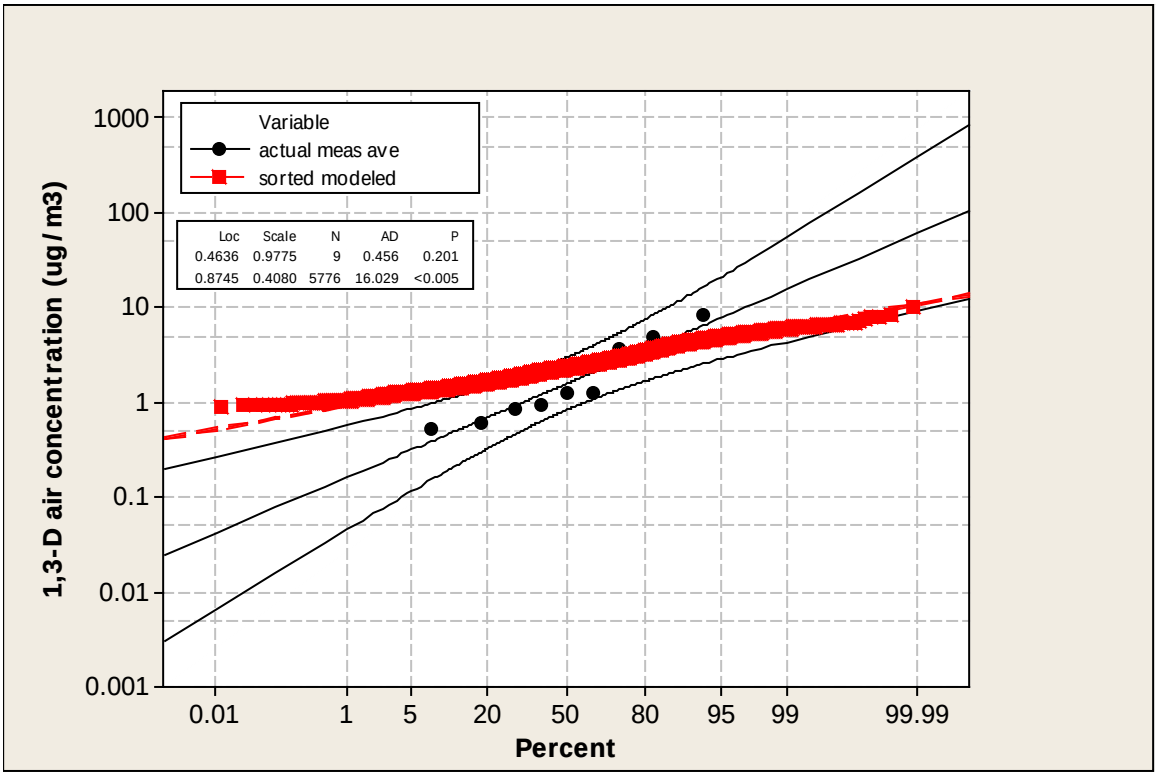


Figure 4 overlays the modeled 1,3-D air concentrations on the estimated measured 1,3-D air concentration probability distribution. The axes are transposed to make visual comparison between the two distributions easier. The same confidence intervals shown in Figures 2 and 3 are shown in Figure 4. Designating the measured values are the benchmark, the ISCST modeled air concentrations are significantly over estimated below the 50th percentile as illustrated by the many modeled values fall outside the measured air concentrations 95% confidence interval. This is consistent with the Dow findings. Above the 50% percentile the modeled values are lower than the measured values but all the modeled values fall within or at the lower 95% confidence interval on the measured air concentrations distribution. The match between these distributions will likely improve with new model runs after the errors in the stability class assignments and mixing height adjustments are corrected and the ISCST model is rerun using the validation scenario inputs with the corrected weather file.

Figure 4. Comparison of the estimated distribution of the measured 1,3-D air concentrations (ug/m^3) with estimated distribution of the SOFEA2 validation scenario modeled 1,3-D air concentrations (ug/m^3).



Conclusions:

The SOFEA2 model cannot be conclusively evaluated due to the following three factors:

1) The atmospheric stability classes assigned for many hours are in error.
2) The mixing height adjustment for many hours are in error.
3) The SOFEA2 model would not successfully perform the post processing of the ISCST modeled Merced validation scenario air concentrations. The SOFEA2 model bombed when attempting to conduct the post processing. As a result, none of the ISCST output files were closed and were lost.

All three of these factors must be fixed before DPR can move forward with a decision whether or not to use the SOFEA2 in house.

Preliminary analysis of the ISCST Merced validation scenario results indicates that if the three issues above are fixed the SOFEA2 model will likely produce modeled air concentrations that reflect the magnitude of the air concentrations measured by the 9 air samplers in the center of the 9 townships over the 14.5 month averaging period. The 72-hour and annual averages will be examined once the next version of SOFEA2 and the corrected Merced weather file with adjusted mixing heights are submitted.

The SOFEA2 model must run easily for scenarios other than those submitted by DOW and without any other significant issues before DPR can consider using it as a modeling tool.

References:

USEPA. 2005. Revision of the guideline on air quality models: Adoption of a preferred general purpose (flat and complex terrain) dispersion model and other revisions. Final Rule. Federal Register. Part III. Environmental Protection Agency. 40 CFP Part 51. Vol. 70, No 216. November 9, 2005. Rules and Regulations.

Johnson, B. 2014. Evaluation Report – Pesticide. Dated 6/23/2014.

Johnson, B., Barry, T. and Wofford, P. 1999. Workbook for Gaussian Modeling Analysis of Air Concentration Measurements. Report No. EH 99-03. Sacramento, CA: Environmental Monitoring Branch, Department of Pesticide Regulation, California Environmental Protection Agency.
http://www.cdpr.ca.gov/docs/empm/pubs/ehapreps/eh9903.pdf

Pasquill, F., 1974. Atmospheric Diffusion, 2nd Edition. John Wiley and Sons, New York, NY

Pratt, G.C., C.Y. Wu, D. Bock, J.L. Adgate, G. Ramachandran, T.H. Stock, M. Morandi, and K. Sexton. 2004. Comparing air dispersion model predictions with measured concentrations of VOC's in urban communities. Environmental Science and Technology Vol 38(7): 1949-59.

Rotondaro, A. and I. van Wesenbeeck. 2012. Montoring of Cis- and Trans- 1,3-Dichlorpopropene in air in 9 high 1,3-Dichlorpopropene use townships Merced County, California. Regulatory Sciences and Government Affairs – Indianapolis Lab. Dow AgroSciences LLC. 9330 Zionville Road, Indianapolis, Indiana 46268-1054. Data volume 50046-0220 parts 1 and 2.

Schnelle, K.B. and P.R. Dey. 2000. Atmospheric dispersion modeling compliance guide. McGraw Hill.

Turner, B. 1994. Workbook of atmospheric dispersion estimates. Second edition. Lewis Publishers. CRC Press, Inc. Boca Raton, FL.

Zanetti, P. 1990. Air pollution modeling. Theroies, computational methods and available software. Van Nostrand Reinhold. New York, New York.

APPENDIX X. Toxicology data review summary for 1,3-D: (http://www.cdpr.ca.gov/docs/risk/toxsums/pdfs/573.pdf)

CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
HUMAN HEALTH ASSESSMENT BRANCH

SUMMARY OF TOXICOLOGY DATA
1,3-DICHLOROPROPENE (TELONE II)

Chemical Code # 000573 Document Processing Number (DPN) # 50046
SB 950 # 137
Original date: August 18, 1986
Revisions: 4/16/87, 7/18/88, 5/23/89, 4/27/90, 6/1/90, 6/15/94, 8/10/94, 5/01/96, 3/11/97, 9/23/99, 3/30/2015, and July 1, 2015

DATA GAP STATUS

Chronic toxicity, rat:	No data gap, possible adverse effects
Chronic toxicity, dog:	No data gap, possible adverse effects
Oncogenicity, rat:	No data gap, possible adverse effects
Oncogenicity, mouse:	No data gap, possible adverse effects
Reproduction, rat:	No data gap, no adverse effects
Developmental toxicity, rat:	No data gap, no adverse effects
Developmental toxicity, rabbit:	No data gap, no adverse effects
Gene mutation:	No data gap, possible adverse effects
Chromosome effects:	No data gap, no adverse effects
DNA damage:	No data gap, possible adverse effects
Neurotoxicity:	Hen neurotoxicity studies are not required at this time

Toxicology one-liners are attached.

All relevant record numbers indexed as July 1, 2015 were examined, including acute studies on technical active ingredient. This includes record numbers up to 283298 (Document No. 50046-0223), as well as relevant older records of the 900,000+ series. Aldous, July 1, 2015.

In the 1-liners below: ** indicates an acceptable study, **Bold face** indicates a possible adverse effect, and ## indicates a study on file but not yet reviewed.

Charles N Aldous July 1, 2015

Leung 7/1/2015

The original Summary was prepared by F. Martz. Revisions in 1987 to 1990 were by J. Gee. Subsequent revisions were by C. Aldous (latest revision being July 1, 2015).

See also "Guidance for the Reregistration of Pesticide Products (Reregistration Standard) Containing 1,3-Dichloropropene (Telone II) as the Active Ingredient," US EPA, 9/18/86, DPR Record # 050620. The position of EPA (1986) was that if significant residues were found, oral studies would be required in addition to existing inhalation studies. This appears to explain the presence of recent dietary chronic studies in rat, mouse, and dog, even though acceptable inhalation studies were previously performed in the rodents. Document No. 50046-116 contains a chapter from the 1997 U.S. EPA RED on 1,3-dichloropropene. Gee, 5/23/89, updated by Aldous, 4/23/96.

NOTE: The following symbols may be used in the Table of Contents which follows:
** = data adequately address FIFRA requirement
† = study(ies) flagged as "possible adverse effect"
(N/A) = study type not currently required

This record contains summaries of studies. Individual worksheets may be useful for detailed assessment.

Table of Contents

METABOLISM AND PHARMACOKINETICS

50046-052 113124 Waechter, J. M., K. A. Brzak, L. P. McCarty, M. A. LaPack, and P. J. Brownson, "1,3-Dichloropropene (Telone II Soil Fumigant) inhalation pharmacokinetics and metabolism in human volunteers," The Dow Chemical Company, Midland, MI, Feb. 12, 1992, Laboratory Study # M-003993-023. Six male volunteers were exposed by inhalation for 6 hours to 1 ppm of *cis*/*trans*-1,3-dichloropropene (1,3-D, Telone II Soil Fumigant), consisting of 50.6% *cis*-1,3-dichloropropene and 45.2% *trans*-1,3-dichloropropene. Investigators assessed concentrations of the above 2 isomers in exhaled air and blood during and after exposure, as well as urinary excretion of two key conjugation products: *cis* and *trans* N-acetyl cysteine (NAC). The respiratory uptake was about 80% for both isomers (based on comparison of chamber air

with exhaled air). There was a rapid "plateau" of concentrations of both isomers in exhaled air in most subjects during exposure, followed by a rapid decrease below detection after termination of exposure in most cases. Subjects absorbed estimated total amounts of 4.2 mg of *trans* and 4.6 mg of *cis* isomers. For technical reasons, only 5 subjects provided blood samples throughout the exposure and post-exposure period. Blood concentrations averaged about 0.7 ng/g for *cis* and about 1.3 ng/g for *trans* Telone, respectively, for most samplings during the exposure period. These levels were about twice the respective limits of detections for respective isomers. For the *cis* isomer, 3 subjects showed no systematic increase between the earlier samplings (10-minutes to 3-hrs) and the 6-hr sampling (end of exposure). The other 2 subjects had a 2- to 3-fold increase of *cis* isomer at 6 hrs compared to the first 3 hours. For the *trans* isomer, all subjects showed a 1.5- to 3-fold increase at 6 hrs compared to the first 3 hours. For both isomers, inter-subject differences were minor for 4 subjects for the first 3 treatment hours. One subject had mostly levels below quantification limits during the exposure phase. One subject had maximal (6-hr) blood levels of both isomers which were markedly higher (up to about 3x) compared to the other subjects, suggesting a meaningful inter-subject variation. Post-exposure blood concentrations in most subjects were generally below detection limits or were marginally quantifiable levels, with occasional detectable levels in some subjects for at least 4 hrs for one or both isomers. The subject with the highest concentrations of both isomers at the final treatment phase assessment still had detectable levels of both isomers 4 hrs into the recovery period. Total urinary elimination by assayed NAC's was quite variable: *cis*-1,3-NAC excretion varied from 2.3 to 5.8 mg, whereas the much lower amounts of *trans* -1,3-D-NAC ranged from 0.4 to 1.9 mg. About 44% of the mass of those conjugates is attributable to parent Telone. Thus, although the NAC conjugates constituted large portions of absorbed Telone II, particularly for the *cis* isomer, fate of much of absorbed Telone II was not addressed in this study. Initial phase half-lives for urinary excretion of *cis* and *trans* NAC's were 4.2 and 3.2 hrs, respectively. Terminal phase half-lives were 12.3 and 17.1 hrs, respectively. This report was first examined by J. Sanborn of the Worker Health and Safety Branch of DPR, whose review described study methodology and key findings. Sanborn noted that although the NAC's are suitable indicators of exposure at the relatively high levels tested, sensitivity of analysis at that time (1992) was insufficient to assess the much lower anticipated exposure levels of field workers and particularly of bystanders. Study is supplementary by design, clearly showing the usefulness of NAC metabolites at high dose levels in persons with fully-functioning GSH conjugation capacity. This subsequent review by C. Aldous (Jan. 6, 2015) contains additional discussion and a table.

50046-120 162473 Stott, W. T., J. R. Gilbert, R. J. McGuirk, K. A. Brzak, M. D. Dryzga, and M. J. Bartels, "Bioavailability of microencapsulated Telone*II Soil Fumigant in Fischer 344 rats", The Dow Chemical Co., Midland, 8/21/96. Laboratory Project Study ID # M-003993-027. Female rats received 25 mg/kg each of microencapsulated (Midwest Research Institute) and neat Telone by gavage in corn oil, prior to sampling the blood for cis and trans isomers over the course of an hour. ^{13}C-telone was used for the neat treatment, whereas the microencapsulated application used the common isotope (^{12}C-telone), allowing investigators to distinguish between modalities of exposure by subsequent mass spectrometry. Microencapsulated Telone was quite stable in corn oil, yet gave peak blood concentrations within minutes after gavage administration. Microencapsulated cis- or trans- 1,3-dichloropropene was absorbed as quickly or more quickly than neat administrations of the same isomers. Urinary excretion half-lives were determined.

Thus microencapsulation is a viable technique for oral administration. Aldous, 9/7/99 (no worksheet).

50046-111 161853 Exact duplicate of 50046-120 162473, above.

50046-026 057488 Stott, W. T., and P. E. Kastl, "Inhalation pharmacokinetics of Technical Grade 1,3-Dichloropropene in rats," The Dow Chemical Company, Midland, MI, Aug. 1, 1985. This unpublished report is virtually identical to the publication: *Toxicology and Applied Pharmacology* publication: Stott, W. T. and P. E. Kastl, "Inhalation pharmacokinetics of Technical Grade 1,3-Dichloropropene in rats," *Toxicology and Applied Pharmacology* 85, 332-341 (1986). Anesthetized male F344 rats were exposed to 1,3-dichloropropene, 92.1% purity (49.3% *cis* and 42.3% *trans*: Lot AGR 204046) by nose-only inhalation exposure at 30, 90, 300, or 900 ppm for 3 hrs for most experiments. Based on breathing rate, tidal volume, and measurements of dichloropropene (DCP) levels in air entering and leaving the head-only exposure space, estimated absorbed dose was 82% of inhaled DCP for 30 ppm exposure, and 62-65% for 90-900 ppm. For blood analysis of *cis* and *trans* DCP, investigators repeatedly sampled blood from indwelling jugular cannulae. At 30-90 ppm, blood levels at 1, 2, and 3 hours of exposure were constant, followed by rapid declines at the end of exposure (especially rapid for *cis* isomer). At 300 ppm and particularly at 900 ppm, blood levels rose markedly from hour to hour during the exposure period. Estimated phase 1 elimination half-lives for *cis* DCP were 3-5 min at 30 to 300 ppm, and 14 min for 900 ppm. Corresponding elimination half-lives for *trans* DCP were about 5 min for lower dose levels, and 27 min for 900 ppm. *Trans* DCP, although not as abundant in the technical as the *cis* isomer, was consistently more abundant in blood during exposure, and was more slowly cleared after cessation of dosing. Blood plateau levels at 30 ppm were 0.085 and 0.12 µg/ml for *cis* and *trans* DCP, respectively. The most profound difference in blood concentrations of isomers at termination of exposure was in 300 ppm rats, where *trans* isomer was over twice the concentration of *cis* isomer. Investigators determined the proportion of absorption in upper and lower respiratory tract (URT and LRT, respectively) after sectioning and catheterizing the tracheae of anesthetized rats. LRT absorption was assessed by analyzing inhaled and exhaled gasses in the isolated caudal portion of the endotracheal tube. URT uptake was estimated by examining input and output gas DCP in a direct unilateral flow model. Sums of LRT and URT uptakes were compared against absorbed dose in similarly anesthetized rats with intact respiratory tracts. These tests evaluated 90 ppm and 150 ppm DCP test atmospheres only. Investigators determined that 73-79% of total absorbed DCP was absorbed in the LRT, the balance of absorbed DCP in the cannulated rats being absorbed in the URT. The sums of these two isolated respiratory tract uptake estimates were acceptably close to measured absorption in the anesthetized intact rat. When expressed as percent of theoretical uptake, LRT and nose-only intact rats absorbed about 50% of available DCP. About 11-16% of dose was absorbed in the URT. Respiratory frequency and tidal volume were assessed with a pressure transducer designed to measure pressure changes in the head-only space. There was a consistent decrease in respiratory rate (breaths/min) with increasing dose, with no consistent change in tidal volume, hence there was a decrease in respiratory minute volume at 300 to 900 ppm. A combination of reduced respiratory minute volume and reduced percent of dose absorbed at the highest dose resulted in a reduction of uptake/(exposure concentration) to less than 50% at 900 ppm compared to 30 ppm rats. Investigators estimated tissue non-protein sulfhydryl (NPSH) content in homogenates of liver, kidney, or lung immediately after dosing with 0 or 90 ppm DCP (tissue

proteins were removed by precipitation with trichloroacetic acid or *m*-phosphoric acid). Liver and kidney NPSH contents were statistically significantly reduced by DCP: 31% reduction of NPSH in kidney and 41% decrease in liver (no NPSH effect in lung). Aldous, Jan. 5, 2015.

50046-0216 282090 Waechter, J. M. and P. E. Kastl, "1,3-Dichloropropene: Pharmacokinetics and metabolism in Fischer 344 rats following repeated oral administration," The Dow Chemical Company, Midland, MI, 12/23/88. Laboratory Study # K-6409-(13). Five rats/sex were dosed daily with 5 mg/kg 1,3-dichloropropene (1,3-D), (54.2% *cis* and 45.8% *trans*), by gavage in corn oil (5 ml/kg) for 14 consecutive days. On day 15, rats were fasted for 8 hrs prior to dosing with 5 mg/kg of *cis/trans*-1,3-dichloropropene, uniformly labeled with ^{14}C, which assayed prior to dosing at 96.3% 1,3-D (53.5% *cis* and 43.0% *trans*). Two additional fasted rats/sex, which had no prior 1,3-D treatment, were treated with the same labeled material. Urine, feces, exhaled CO_2, other expired volatiles, and tissue levels were assessed. Sacrifice was 48 hrs after labeled treatment. Chromatic resolution was not sufficient to routinely separate the *cis* and *trans* isomers of mercapturic acid conjugates, and also not sufficient to separate subsequent sulfoxide and sulfone products from one another. Approximately 62-65% of administered dose was found in urine, 26% in exhaled CO_2, 5% in feces, and 4-6% in tissues and carcass, with no observed sex difference. Disposition in the 2/sex non-pre-treated rats was quite similar. At 48 hrs after dosing of pre-treated rats, highest concentrations of label (about 1 µg equivalent/g tissue) were in bladder and forestomach. Lowest concentrations were in brain and fat (less than 0.1 µg equivalent/g tissue), with about 0.2 to 0.4 µg equivalent/g in other tissues. Urinary excretion of the *cis/trans* N-acetyl cysteine conjugates comprised 26-28% of administered dose. The combined close-eluting sulfoxide and sulfone residues comprised about 14% of administered dose. No other components were characterized. Most excretion occurred within the first 12 hours. There were no clear differences in disposition between sexes or resulting from 14-day pre-treatment vs. naïve rats. This is a valid supplemental study. Aldous, 1/23/15.

50046-058 115135 Stott, W. T., "Implications of the results of a recent human pharmacokinetics study upon interspecies extrapolation of risk," (3-page letter). Stott determined by extrapolation from a human volunteer study [Waechter et al., 1992, Record No. 113124] and from a rat pharmacokinetics study Stott, W. T., and P. E. Kastl, 1986 (published), or DPR unpublished version in Record No. 057488] that humans attain a lower systemic dose than do rats following equivalent inhalation exposure. Stott also concluded [evidently from study by Waechter et al., 1992] that humans rapidly clear Telone from the blood ($T_{1/2}$ probably less than 10 min). Also, humans evidently excrete Telone as conjugates (largely mercapturates, and likely also sulfoxides) to a large degree, perhaps more than do rodents. This letter was not reviewable and thus has no worksheet. Aldous, Jan. 9, 2015.

50046-060 118048 Stott, W. T. and L. L. Calhoun, "Implications of the results of a recent human pharmacokinetics study upon interspecies extrapolation of risk," (3-page letter). This provides a few updates to the above record 50046-058 115135 by Stott (identical titles in both letters), and makes a strong statement that (1) default animal-to-human scaling factors to address metabolic capacity for humans are not appropriate, consideration the data available for human subjects already account for physiological differences, and (2) a default potency calculation based on the mouse and not the rat already leads to excessively conservative outcomes. This letter was not reviewable and thus has no worksheet. Aldous, Jan. 12, 2015.

50046-0015 932855 Dietz, F. K., E. A. Hermann, and J. C. Ramsey, "The Pharmacokinetics of ^{14}C-1,3-dichloropropene in rats and mice following oral administration," Toxicology Research Laboratory, Dow Chemical U.S.A., Midland, MI. This appears to be an abstract for a professional meeting, with no date given, however this abstract pre-dates the accompanying 9/22/1983 letter in the volume. Investigators reported urinary excretion of an oral dose of Telone was 51-61% of dose in rats, and 63-79% in mice over the 48-hr study period. Feces accounted for 18% and 15% of dose in rats and mice, respectively. Expired CO_2 comprised 6% and 14% of dose in rats and mice. About 2-6% of dose remained in carcasses in either species at 48 hrs. The major identified metabolite was N-acetyl-S-(3-chloroprop-2-enyl) cysteine. A second major metabolite appeared to be a sulfoxide or sulfone of the latter. This abstract was not reviewable and thus has no worksheet. Aldous, Jan. 12, 2015.

50046-0015 932856 Dittenber, D. A, H. D. Kirk, and J. C. Ramsey, "Non-protein sulfhydryl content and macromolecular binding in rats and mice following oral administration of 1,3-dichloropropene," Toxicology Research Laboratory, Dow Chemical U.S.A., Midland, MI. This appears to be an abstract for a professional meeting, with no date given, however this abstract pre-dates the accompanying 9/22/1983 letter in the volume. Investigators administered oral doses of Telone to male F344 rats and to B6C3F1 mice at 0 to 100 mg/kg. After 2 hrs, animals were sacrificed for collection of forestomach, glandular stomach, liver, kidneys, and bladder. Depletion of non- protein sulfhydryl was highest in forestomach (to as low as 17-27% of control levels at 100 mg/kg). Also, tissue binding of label was highest in forestomach and glandular stomach (of tissues sampled). Investigators concluded that these tissues in particular would be particularly sensitive to Telone at high dose levels. This abstract was not reviewable and thus has no worksheet. Aldous, Jan. 12, 2015.

GUIDELINE ACUTE STUDIES ON ACTIVE INGREDIENT

Acute oral toxicity, rat ** (related non-guideline studies included)

50046-032; 62068; Telone II Soil Fumigant: Acute Oral Toxicity Study In Fischer 344 Rats; Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, Dow Chemical Co., Midland MI, Project ID HET M-003993-017A, 2/9/87; (Telone II Soil Fumigant); Dose levels 100, 500, and 1000 mg/kg of a 10% (v/v) solution of the test material in corn oil; 5 animals/sex/dose; Clinical Observations-lethargy, diarrhea, lacrimation, palpebral closure, labored respiration, facial soiling and/or rough hair coat in some animals of all dose groups; LD50 (M) = 300 mg/kg, LD50 (F) = 224 mg/kg; CAT II; Acceptable; JSB, 5/2/88

50046-0223 283294 Toyoshima, S., R. Sato, and S. Sato, "The acute toxicity test of Telone* II in mice," Japan Experimental Medical Research Institute Co., LTD, 6/30/78. This is a brief report of an old study, using 92% purity Telone II in JCL:ICR mice. Reported LD_{50} by gavage (corn oil vehicle) was 640 mg/kg in males and also 640 mg/kg in females. Reported LD_{50} by subcutaneous treatment (corn oil vehicle) was 330 mg/kg in males and also 345 mg/kg in females. "Percutaneous" exposure (fixed application of 1 ml/kg over 3 x 2 cm^2 of skin) caused no deaths and did not elicit any findings of toxicity, hence $LD_{50} > 1.211$ mg/kg in both sexes. Not acceptable and not upgradeable, as test article was unlikely to represent modern technical Telone II, and report lacked sufficient detail to be suitable for a worksheet. Aldous, 3/30/15.

50046-0223 283295 Toyoshima, S., R. Sato, and S. Sato, "The acute toxicity test of Telone* II in rats," Japan Experimental Medical Research Institute Co., LTD, 6/30/78. This is a brief report of an old study, using 92% purity Telone II in Wistar rats. Reported LD_{50} by gavage (corn oil vehicle) was 560 mg/kg in males and also 510 mg/kg in females. Reported LD_{50} by subcutaneous treatment (corn oil vehicle) was 400 mg/kg in males and also 366 mg/kg in females. "Percutaneous" exposure (fixed application of 1 ml/kg over 4 x 5 cm^2 of skin) caused no deaths and did not elicit any findings of toxicity, hence LD_{50} > 1.211 mg/kg in both sexes. Not acceptable and not upgradeable, as test article was unlikely to represent modern technical Telone II, and report lacked sufficient detail to be suitable for a worksheet. Aldous, 3/30/15.

Acute dermal toxicity (see note at end of 1-liner)

50046-032; 62069; Telone II Soil Fumigant: Acute Dermal Toxicity Study In New Zealand White Rabbits; Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, Dow Chemical Co., Midland MI, Project ID HET M-003993-017D, 3/18/87; (Telone II Soil Fumigant); Dose levels 200 and 1000 mg/kg; 5 animals/sex/dose; 24 hour exposure period, occluded patch; Clinical Observations-restless, squealing, lethargic, diarrhea, anorexia, labored breathing; Dermal Irritation- all animals had skin and/or subcutaneous tissue irritation at the dermal site, subcutaneous, necrosis, erythema, edema, and/or crusts; Report states the LD50 (M/F) = 333 mg/kg which falls into a CAT II. Unacceptable, as guidelines require at least three dose levels to produce test groups with a range of toxic effects and mortality rates; J. Berliner, 5/3/88. NOTE: Current evaluation guidelines would have allowed acceptance of this study, since dose levels identified a toxicity category (Aldous, 6/25/15).

Acute inhalation toxicity, rat **

**50046-032; 62070 "Telone II Soil Fumigant: An Acute Vapor Inhalation Study In Fischer 344 Rats," Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, Dow Chemical Co., Midland MI, Project ID HET M-003993-018, 4/30/87. Telone II Soil Fumigant, Lot # TB-860825-5, was administered at nominal/analytical 1076/1035, 946/855, 820/775 ppm to 5 animals/sex/dose for a 4 hour exposure period. Clinical observations were tremors, convulsions, salivation, lacrimation, diarrhea, lethargy, and other signs of altered central nervous system function. LC50 (M) was between 1035 and 855 ppm, LC50 (F)= 904 ppm. Conversion from ppm to mg/l: LC50 (M) between 4.688 mg/l and 3.873 mg/l,LC50 (F)= 4.095 mg/l. Necropsy: clear facial soiling and/or hemorrhages in multiple lung lobes. Category III; Acceptable; J. Berliner, 5/3/88.

50046-0221 282875 Cracknell, S., G. Jackson, and C. Hardy, "Telone* II (1,3-dichloropropene) acute inhalation study in rats, 4 hour exposure," Huntingdon Research Centre, Ltd., 3/25/87. Report designation: DWC/484. Five Wistar-derived rats/sex/group were dosed for 4 hours with Telone II (Lot 071.3054/062), 98.4% purity, by whole-body inhalation at 0, 1.62, 2.64, 2.70, or 3.07 mg/L (assayed mean concentrations). Rats were observed at 6 intervals during exposure, and daily during a 14-day post-dose observation period. All rats survived except for 3/5 males and 3/5 females at 3.07 mg/L. All male groups showed a body weight loss at day 1 weighing, followed by normal subsequent gains in survivors. Body weights were unaffected in 1.62 mg/L females, but other female groups suffered body weight losses on day 1, followed by normal weight gain patterns thereafter. An exception was that the surviving 3.07 mg/L females appeared

to require an additional 5 days for weight gains to normalize. Common signs at the lowest dose (1.62 mg/L) during exposure were partial closing of eyes, slow respiratory rate, and irregular respiratory movements during exposure in all rats; and hunched posture, restlessness, exaggerated respiratory movements, and pawing behavior in some rats. Clinical signs for 1.62 mg/L were normal by the day after treatment, whereas symptoms continued for up to 5 days at 3.07 mg/L. LC_{50} was thus between 2.70 and 3.07 mg/L in both sexes. Study is not acceptable, but upgradeable on submission of a standard curve and sample calculation to validate the reported assayed test atmosphere content. Aldous, April 6, 2015.

50046-0217 282091 Nitschke, K. D., J. W. Crissman, and D. J. Schuetz, "*cis*-1,3-Dichloropropene: acute inhalation study with Fischer 344 rats," The Dow Chemical Company, Midland, MI, 9/27/90. Laboratory Study # K-020256-005. Groups of 5 rats/sex were dosed by whole-body inhalation for 4 hours with time-weighted average concentrations of 573, 771, or 1020 ppm of cis-1,3-dichloropropene. Test article was 95.6% cis-1,3-dichloropropene, with minor amounts of two assayed components: 1.5% trans-1,3-dichloropropene, and 0.2% 1,2-dichloropropane. All rats died at 1020 ppm during or shortly after exposure. All males died at 771 ppm, two of which did not survive more than 30 minutes after the end of dosing. Also 3/5 females died at 771 ppm. All rats survived at 573 ppm. Clinical signs of labored breathing and "eyelids closed" were each observed in 1/sex at 573 ppm on the day of exposure, with no clinical signs in 573 ppm rats after the day of treatment. At 573 ppm, body weights were remarkably diminished on weighing days 2 and 4, with substantial body weight recovery evident by day 8. Gross examinations revealed unilateral opacity in two high dose males, visceral congestion in four of five 771 ppm males, liver and lung congestion in all high dose males, hydrothorax in two 771 ppm males, corneal opacities in the majority of 1020 ppm females, liver and lung congestion in all high dose females, and lung edema in one mid-dose female. All 573 ppm rats were grossly normal at termination. Nominal LC_{50} was estimated to be 670 ppm and 744 ppm for males and females, respectively. For males, the most sensitive gender, mass/volume units of LC_{50} are 3.04 mg/L. This is a supplementary study on a test article enriched in the *cis* isomer. Aldous, 2/2/15.

50046-0206 273926 "Acute inhalation reference concentration assessment for 1,3-dichloropropene," 07/18/2013. This is a discussion on acute inhalation reference concentration assessment by Maier, A., L. Haber, and A. Parker. A DPR review of this submission by L. M. Hall is included in this volume.

50046-0223 283297 Stevenson, D. E., "Toxicity of soil fumigants: acute inhalation toxicity of 1,3-dichloropropene," Shell Toxicology Laboratory (Tunstall), March, 1977. [Technical was 51% cis and 43.4% trans 1,3-dichloropropene, with 1% epichlorohydrin.] Male and female Wistar rats were dosed by whole-body inhalation for 4 hours to establish an LC_{50}. Calculated LC_{50} was 729 ppm for both sexes. This very brief (3-page) summary report on an outdated technical formulation is not suitable for a DPR worksheet. Aldous, 3/30/15.

50046-0223 283298 Yakel, H. O., and R. J. Kociba, "Acute inhalation toxicity of M-3993 (Telone II) in rats," The Dow Chemical Co., Midland, MI, 6/17/77. Test article was "M-3993," composed of 92% 1,3-dichloropropene and 8% inerts (not further described). Ten Spartan Sprague-Dawley rats/sex were dosed for 1 hour at 5.2 mg/L (nominal concentration), with reported mean diameter of 2.96 microns (99% of particles < 6 micron diameter). "Slight

transitory eye irritation" (not further described) was observed during exposure. No behavioral changes were noted, and autopsy findings after 2 weeks were normal. This brief summary report, not a standard design, too brief for thorough review, and likely on an outdated technical formulation, is not suitable for a DPR worksheet. Aldous, 3/20/15.

50046-0223 283296 Landry, T. D., and S. M. Krieger, "Telone II soil fumigant: 1-hour acute nose-only inhalation toxicity study in Fischer 344 rats," The Dow Chemical Co., Midland, MI, Jan. 6, 2003. Five male rats were dosed once as indicated at 14000 ppm Telone II [97.5% a.i., (cis and trans)]. All rats survived the 1-hr exposure, but one died within an hour after treatment, and all were dead by the next day. Major clinical signs included decreased muscle tone, decreased resistance to removal, decreased extensor thrust, decreased reactivity to stimuli, inability to walk, eyelids partially closed, cold to touch, slow and labored respiration, and soiled fur (mostly urine). Mottled lungs and serosanguineous muzzle soiling were the most common gross pathology findings. This was a valid supplementary study, but not of standard design and featuring only one very high dose, hence no DPR worksheet is appropriate. Aldous, 3/20/15.

Primary eye irritation, rabbit **

50046-032; 62071; Telone II Soil Fumigant: Primary Eye Irritation Study In New Zealand White Rabbits; Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, Dow Chemical Co., Midland MI, Project No. HET M-003993-017C, 1/13/87; (Telone II Soil Fumigant); Dose level 0.1 ml; 4M and 2F rabbits with unwashed eyes; left eye remained untreated and served as a control; 7 Day Readings (Unwashed) positive effects only: 1/6 animals score of 2 for redness, and 1/6 animals score of 1 for corneal opacity
14 Day Readings (Unwashed): 6/6 animals score of 0 for redness, chemosis, discharge, and corneal opacity; CAT II; Acceptable; JSB, 5/3/88

Primary dermal irritation **

50046-032; 62072; Telone II Soil Fumigant: Primary Dermal Irritation Study In New Zealand White Rabbits; Mammalian and Environmental Toxicology Research Laboratory, Health and Environmental Sciences, Dow Chemical Co., Midland MI, Project ID HET M-003993-017B, 1/7/87; (Telone II Soil Fumigant); Dose level 0.5 ml; 4 hour exposure period, occluded patch; 6 animals used; 72 Hour Readings (Intact): Erythema- 5/6 animals score of 2, 1/6 animals score of 1. Edema- 2/6 animals score of 2, 2/6 animals score of 1, and 2/6 animals score of 0; CAT III; Acceptable; JSB, 5/3/88

Dermal sensitization ** †

****50046-0032 062073** Jeffrey, M. M., "Telone II Soil Fumigant: Dermal sensitization potential in the Hartley albino guinea pig," The Dow Chemical Company, Midland, MI. March 12, 1987. Laboratory Study # HET M-003993-017E. Ten Hartley albino guinea pigs/group were dosed with either 0.4 ml 0.1% Telone II in mineral oil, a positive control of 10% DER 331 in DOWANAL DPM (eventually reduced to 5% solution due to erythema during the induction phase), or mineral oil vehicle. The above treatments were undertaken weekly three times to the clipped dorsal skin, secured under dressing for 6 hour periods each time, for the induction phase. Two weeks after the last induction phase, a comparable challenge dose was administered. Treatment sites were examined on the day after dosing for each induction treatment, and 24 and 48 hours after the challenge dose. Induction treatments of Telone II did not cause irritation after the first or second treatment. Four of 10 Telone II animals showed slight erythema after the third

and final induction. At 48 hours after challenge, 8/10 Telone II animals displayed slight erythema, and 1/10 Telone II animals displayed moderate erythema. The overall response of Telone II animals at challenge treatment was thus 9/10. Five of 10 positive controls responded with slight erythema by 24 hours at challenge. Mineral oil animals were uniformly negative for both induction and challenge phases. Telone II is a **sensitizer**. This study is acceptable. Aldous, 1/30/15.

Acute Studies with known outdated technical material

50046-0011 932843 "Acute Toxicological Properties of Experimental Nematicide Formulation M-3993 Containing 1, 3 Dichloropropene," Dow Chemical, Midland, MI, 09/01/1975. This is a report of several acute studies of an old active ingredient containing 1% epichlorohydrin. Data are unlikely to be useful any more. Acute NOEL (rat) was calculated to be 713 mg/kg (M) and 470 mg/kg (F). Eye irritation (rabbits) indicated slight to moderate corneal effects at 24 hrs, with clearing by day 8. Skin irritation methodology (rabbits) was unlike modern studies, and did not follow animals for the requisite time after dosing: some edema was evident at 24 hours. Acute percutaneous absorption in rabbits indicated a "mixed rabbit sexes" NOEL of 504 mg/kg. No DPR worksheet is relevant. Aldous, Feb. 2, 2015.

SUBCHRONIC STUDIES (may include subacute probe studies)

Oral subchronic toxicity, rodent: **

50046-073 126523 Haut, K. T., K. A. Johnson, S. N. Shabrang, and W. T. Stott, "Telone II soil fumigant: 13-week dietary toxicity and 4-week recovery studies in Fischer 344 rats", The Dow Chemical Co., Midland, 1/8/93. Laboratory Project Study ID: M-003993-028. Fischer 344 rats, 20/sex/group, were dosed with 0, 5, 15, 50, or 100 mg/kg/day 1,3-dichloropropene in diet (test article was microencapsulated in spheres composed of an 80%/20% starch/sucrose matrix). All rats were exposed for 13 weeks, at which time 10/sex/group were sacrificed, whereas the remaining 10/sex/group were maintained off treatment for 4 weeks to evaluate recovery. The NOEL for toxic endpoints is 5 mg/kg/day (dose-related non-glandular stomach basal cell hyperplasia at 15 mg/kg/day and above). Small but statistically significant decrements in body weight at 5 to 15 mg/kg/day in males and 15 mg/kg/day in females were plausibly treatment-related, but of unlikely toxicological significance (small degree of change or nearly flat dose-response curve). **No adverse effects. Acceptable. Aldous, 9/21/99.

50046-059; 117681; "Telone II Soil Fumigant: Palatability and Two-Week Dietary Probe Studies in Fischer 344 Rats," K.T. Haut *et al*. Non-guideline; Rat; The Toxicology Research Laboratory, Health and Environmental Sciences, The Dow Chemical Company, Midland, MI; Study No. M-003993-026; 8/3/92. Telone II Microencapsulated (a.i.: 42.7%); five rats/sex per group received 0, 10, 25, 50, 100 mg/kg/day (based on a.i. concentration? in starch/sucrose suspension) for 14 days, dietary; No mortality; Reduced body weight gain and food consumption (50 and 100 mg/kg/day); No treatment-related effect on hematology or ophthalmology; Necropsy: no treatment-related lesions; Histopathology: hyperkeratosis in the nonglandular mucosa of the stomach, and thickened nonglandular mucosa of the stomach (considered to be localized irritant effect): (both findings at 50 and 100 mg/kg/day, also 1/5 males had thickened non-glandular mucosa at 25 ppm). NOEL cannot be established until analytical data on the

dietary preparations are submitted, but provisionally is 10 mg/kg/day for males and 25 mg/kg/day for females. Study is supplemental. (Moore, 7/14/94)

50046-074 126524 Haut, K. T., K. E. Stebbins, S. N. Shabrang, and W. T. Stott, "Telone II soil fumigant: 13-week dietary toxicity study in B6C3F1 mice", The Dow Chemical Co., Midland, 1/8/93. Laboratory Project Study ID: M-003993-029. Mice, 10/sex/group, were dosed with 0, 15, 50, 100, or 175 mg/kg/day 1,3-dichloropropene in diet (test article was microencapsulated in spheres composed of an 80%/20% starch/sucrose matrix). Mice were exposed for 13 weeks in a standard subchronic study design. Decreased size of hepatocytes was observed in all male treatment groups ("very slight" degree in all cases): this was attributed by the authors to a decrease in glycogen content. No comparable findings were reported in females. A NOEL for body weight decrements was 15 mg/kg/day for both sexes. Decreased circulating triglyceride levels were found in 100 to 175 mg/kg/day females. All findings were consistent with reduced nutritional status, thus no target organ toxicity was evident. **Acceptable, with no adverse effects. Aldous, 9/20/99.

Inhalation subchronic toxicity, rodent: † (two valid supplementary studies)

50046-038 071713 "Telone II Soil Fumigant: A 13-Week Inhalation Study in Rats and Mice." (Dow, 11/30/84) Telone II, 90.9%, lot WP-82-1111-56, was given by whole-body inhalation for 6 hours/day, 5 days/week for 13-weeks to 10 Fischer 344 rats per sex and to 10 B6C3F1 mice per sex, at 0, 10, 30, 90 or 150 ppm nominal chamber concentration. Body weights were significantly lower at 90 and 150 ppm in rats, dose-related, from treatment day 3 onward. Mouse body weight decrements were consistently reduced at 150 ppm, and occasionally at 90 ppm. Food consumption was not assessed in either species. The only histopathology findings in **rats** were degeneration in the olfactory epithelium at 150 ppm, and hyperplasia of the respiratory epithelium in both sexes, with severity increased from 90 to 150 ppm. Rat NOEL = 10 ppm based on hyperplasia of the respiratory epithelium in 2/10 males at 30 ppm. **Mice** had degeneration in the olfactory epithelium and hyperplasia of the respiratory epithelium in all observed survivors of both sexes at 90 to 150 ppm, with a clear dose-response for severity in that range. Slight olfactory respiratory metaplasia was also observed in all 150 ppm males and in most 150 ppm females (described as replacement of sensory epithelia with ciliated cells typical of epithelia of non-sensory regions of the respiratory tract). In addition, most female mice also displayed epithelial cell hyperplasia of urinary bladder transitional cells, such that affected areas had remarkably increased cell layers of epithelial cells. Rat NOEL = 10 ppm based on hyperplasia of the respiratory epithelium in 2/10 males at 30 ppm. Mouse NOEL = 30 ppm. Report contains valid supplementary data (Gee, 5/22/89, re-examined for a targeted review by Aldous at request of Risk Assessment staff on 1/21/15).

50046-010 036551 Coate, W. B., "90-Day Inhalation toxicity Study in Rats and Mice: Telone® II." The original Hazleton report was completed on 5/15/79. A key addendum consisting of re-evaluation of nasal cavity histopathology was undertaken for all groups of both species (July 9, 1979). This was not a guideline study, but was intended as range-finder for future studies. Ten rats/sex and 10 mice/sex were exposed by whole-body inhalation to Telone II, "Production Grade," purity unspecified, at 0, 10, 30, or 90 ppm for 6 hours/day, 5 days/week, for 13 weeks (65 exposures). NOEL for female rats is 12 ppm, based on decreased cytoplasmic content and disorganization of cell nuclei in nasal epithelial cells of the nasal septum and dorsal turbinates.

NOEL for male rats is 32 ppm, based on the above histopathology in all 93 ppm rats. There were body weight gain decrements in both sexes at 93 ppm, and some individuals of both sexes of rats at that dose had individual cell necrosis in nasal epithelia. NOEL for mice is 32 ppm, based on body weight gain decrements in both sexes, and on decreased cytoplasmic content in nasal epithelial cells of the nasal septum and dorsal turbinates in females. This is a valid supplementary study, with nasal turbinate histopathology designated as a "possible adverse effect." (F. Martz, 4/29/86. This report was later reviewed to support risk assessments on 7/6/89 by C. Lewis, and on 1/29/15 by C. Aldous).

50046-0013 932848 Parker, C. M., W. B. Coate, and R. W. Voelker, "Subchronic inhalation toxicity of 1,3-dichloropropene/1,2-dichloropropane (D-D) in rats and mice," Journal of Toxicology and Environmental Health 9:899-910 (1982). Test article was an old Shell product comprised principally of *cis*-1,3-D (25%), *trans*-1,3-D (27%), and 1,2-dichloropropane (29%). Investigators reported significantly increased incidence of slight "diffuse hepatocytic enlargement" in male mice, and a marginal increase in female mice, with no notable findings of any kind in rats. Given the difference between this and any contemporary products, this study has no apparent importance. Aldous, Feb. 1, 2015. No worksheet.

Oral subchronic toxicity, non-rodent:

046 075537 "Telone II: 13-Week Dietary Toxicity Study in Beagle Dogs." Quast, J. F., Dow Chemical Company, August 1, 1989. The 3-page letter was submitted as an adverse effects disclosure for microcytic hypochromic anemia in the 13-week study in beagle dogs. Doses were 0, 130, 380 or 1000 ppm with Telone II incorporated in a starch sucrose matrix and administered in the dog chow. The letter contains no data but states the anemia was dose-related. Page 2 of the letter indicates "minimal effect in one dog of each sex" at 130 ppm, presumably for microcytic hypochromic anemia. Some dogs were being maintained after dosing for further study. The final report has not yet been received by CDFA. (Gee, 4/27/90). NOTE: As of 2/2/15, no finished dog subchronic dietary report has been received (C. Aldous).

50046-043 protocol (dated 4/12/89) for the 13-week dietary dog study (see Record No. 075537, above). (Gee, 5/23/89).

Dermal toxicity, 21/28-day or 90-day:

CHRONIC STUDIES

Combined (chronic and oncogenicity), rat

Oral route ** †

****50046-098 140562** Stott, W. T., K. A. Johnson, T. K. Jeffries, K. T. Haut, and S. N. Shabrang, "Telone*II Soil Fumigant: Two-year chronic toxicity/oncogenicity study in Fischer 344 rats". The Toxicology Research Laboratory, The Dow Chemical Co., Midland MI, 8/15/95. Laboratory Project Study ID: M-003993-031. Microencapsulated Telone*II, purity 95.8% 1,3-dichloropropene (50.7% cis/45.1% trans), was admixed with the diet at 0, 2.5, 12.5 or 25 mg/kg/day and fed to 50 F344 rats/sex/group for 24 months. Additional rats (10/sex/group) were

allocated for a 1-yr interim sacrifice. No definitive NOEL is present in this study: hepatocellular eosinophilic foci appear increased in number and/or degree at all dose levels. Characteristic findings at 12.5 to 25 mg/kg/day include forestomach basal cell hyperplasia, reduced body weight, and reduced food consumption. Hepatocellular tumors (primarily adenomas) were significantly elevated in high dose males, and non-significantly elevated in mid-dose males and high dose females. The 1996 DPR review noted an apparent elevation in uterine endometrial stromal polyps in 25 mg/kg/day females, however supplemental data in Document No. 50046-104, Record No. 151747 present historical control data which do not support a treatment effect. A number of other changes (particularly reductions in degree or incidence of normal aging lesions) indicate altered physiology or nutritional status influencing the progress of normal geriatric changes in high dose rats. Study is **acceptable**. Liver tumors are "possible adverse effects". Kishiyama and Aldous, 4/19/96; Aldous, 3/11/97.

50046-072 126522 This 116-page report is the 1-yr interim report of Record # 140562 above. There is no essential new information in this report , hence no worksheet. Aldous, 7/15/99.

F**ollowing is a mechanistic study, primarily applicable to study 50046-098 140562, above.**
Klaunig, J. E., S. C. Gehen, Z. Wang, P. J. Klein, R. Billington, "Mechanism of 1,3-dichloropropene induced rat liver carcinogenesis," ToxSci Advance Access published 10/28/14. Participants were affiliated with Indiana University and Dow Agro Sciences. All test animals were F344 rats, which were pre-treated with dimethylnitrosamine (DEN) twice (at 6 and at 7 weeks of age, 100 mg/kg each time) to initiate pre-neoplastic foci in the liver. Sixteen weeks later, groups of 11 were dosed as follows: (1) 1,3-Dichloropropene (1,3-D): 52.5% *cis*- and 46.7% *trans*-, without stabilizers such as epichlorohydrin; groups of 11 were administered by gavage in corn oil daily for 30 days, for 60 days, or for 30 days followed by 30 days of recovery; (2) phenobarbital (PB), 80 mg/kg/day by gavage in saline; groups were dosed daily for 30 days, for 60 days, or for 30 days followed by 30 days of recovery; or (3) negative controls were treated with corn oil for 30 days or for 60 days. Liver tissues were fixed, embedded, and stained with H&E. Focal liver lesions were also assessed with immunohistochemical techniques targeting placental glutathione S-transferase (GSTP), which is used as a marker for pre-neoplastic lesions, and which is often associated with phenobarbital enhancement of liver tumors. Also, BrdU immunohistochemical techniques (after osmotic pump infusion for 7 days prior to sacrifice) were used to assess cell replication. Investigators also employed a 2-dimensional morphometric analysis to evaluate sizes of focal lesions. Body weights did not vary systematically with treatments. Liver weights were greatly increased in PB groups after 30 days and 60 days, with no significant differences after 30-day recovery. 1,3-D had no influence on liver absolute or relative weights. BrdU labeling index was elevated about 2-fold in GSTP-negative foci at 30 and at 60 days in 1,3-D rats, and at 60 days in PB rats. There were no such changes in GSTP-positive foci. Total numbers of GSTP-positive foci were unchanged with 1,3-D. In contrast, PB elicited an increase in GSTP-positive foci correlated with exposure duration between 30 and 60-day exposures, with a significant residual elevation of GSTP-positive foci in PB rats after the recovery phase. GSTP-negative foci were statistically significantly increased in number following 1,3-D treatment for 30 or 60 days, with a modest elevation remaining after recovery. PB did not have a consistent effect on GSTP-negative foci. Volume of GSTP-positive foci (assessed by 2-dimensional analyses) was quite variable, and the only statistically significant finding was an increased total volume of foci in the 60-day PB group. Volumes of GSTP-

negative foci were increased in 1,3-D groups at 30 and 60 days. There was also a comparable increase in volume of GSTP-negative foci in PB rats at 60 days. An assessment of numbers and size class of GSTP-positive foci found a progressive increase in size and number of the PB groups compared to the other two groups from the 30-day to 60-day exposure intervals. A small but visually evident increase in focal size class was evident in the recovery PB rats. An assessment of numbers and size class of GSTP-negative foci found 1,3-D foci to have increased abundance among small to medium-sized foci after 30 and at 60 days exposure, with no residual 1,3-D effect after the recovery phase. PB rats treated for 60 days had slightly elevated numbers of relatively large GSTP-negative foci compared to other groups. This report provides useful data, showing that 1,3-D is capable of promoting previously-induced GSTP-negative foci with an enhancement of DNA synthesis, without eliciting the GSTP-positive foci that are characteristic of PB exposure, and which are also associated with non-genotoxic tumor promotion. Examined tissues in this study returned to normal appearance and function upon removal of 1,3-D exposure. Aldous, 2/2/15.

50046-010 036552 "Toxicology and Carcinogenesis Studies of Telone II in F344/N Rats and B6C3F1 Mice." (NTP Technical Report Series No. 269, Frederick Cancer Research Center, 5/85). **This 1-liner summarizes results in rats**. Telone II (1,3-dichloropropene, 87.5% pure, with epichlorohydrin as stabilizer) was administered to 52/sex/group at 0, 25, or 50 mg/kg by oral gavage "3 times a week" for the lifetime study. An additional 25/sex/group constituted satellite groups for general toxicity evaluation, with sacrifices at 9, 16, 21, 24 and 27 months of 5/sex/group. Study is scientifically valid but unacceptable and not upgradeable because it was not designed to address FIFRA guidelines. **Forestomach** was a target organ, with primary non-neoplastic lesion of basal cell hyperplasia incidence (out of 52) in control through high dose males of 1, 3, and 9, and in females of 0, 0, and 12. Respective forestomach squamous cell papilloma incidences in the lifetime study were 1, 1, and 9 for males, and 0, 2, and 3 for females. Respective forestomach squamous cell carcinoma incidences were 0, 0, and 4 in males, with no squamous cell carcinomas in females. **Liver** did not show non-neoplastic change with dose, but tumor incidence appeared to be meaningfully elevated in males. Incidence of "neoplastic nodule" in control through high dose lifetime study males was 1, 6, and 7; and for corresponding females was 6, 6, and 10. Incidence of hepatocellular carcinoma was 0, 0, and 1 in respective males, with no carcinomas in females. Forestomach pathology, including tumors, is a "possible adverse effect." Study was initially reviewed on 1/16/86 by Martz. Aldous made edits without a new worksheet on 2/3/15.

Inhalation route †

****50046-0031 060677** Lomax, L. G., L. L. Calhoun, W. T. Stott, and L. E. Frauson, "Telone* II soil fumigant: 2-year inhalation chronic toxicity-oncogenicity study in rats," The Dow Chemical Company, Midland, MI, 7/13/87. Laboratory Study # M-003993-009R. Test article was 1,3-Dichloropropene, 92.1% (cis 49.5% and trans, 42.6%) stabilized with soybean oil. For the lifetime study, 50 F344 rats/sex/group were exposed by inhalation 6 hours/day, 5 days/week for 2 years - whole body exposure - at 0, 5, 20 or 60 ppm nominal. Histopathology of interest was mainly limited to nasal olfactory epithelium (decreased thickness, mainly bilateral; and erosions, most commonly bilateral). In some cases, the submucosa underlying the olfactory epithelium showed slight to moderate fibrosis. There were no treatment-related tumors in any tissue. Except for one 20 ppm male, the nasal histopathology was limited to 60 ppm rats, with incidences highest in males. Thus NOEL for males = 5 ppm (due to the one individual with

nasal pathology similar to common responses in 60 ppm rats). NOEL for females = 20 ppm. Body weights were modestly but statistically significantly reduced in 60 ppm males and females for the approximately the first year of the study. ACCEPTABLE with possible adverse chronic effects (above nasal olfactory epithelial responses). (Gee, 7/11/88, re-evaluation by Aldous on 1/26/15 at the request of Risk Assessment Group).

50046- 005 036218 Stott, W. T., L. G. Lomax, L. L. Calhoun, and J. F. Quast, , "Telone* II soil fumigant: 2-year inhalation chronic toxicity-oncogenicity study in rats – **interim report: 6- and 12-month interim sacrifice of rats**," The Dow Chemical Company, Midland, MI, 9/27/85. This was the satellite study to the rat combined study (DPR Document No. 50046-0031, Record No. 060677, above). Rats were dosed in the same chambers concurrently with that lifetime study, with groups of 10/sex/group designated for 6- and 12-month sacrifices. At respective terminations, rats were evaluated for hematology, clinical chemistry, urinalysis, and histopathology. Except for modest but consistent body weight decrements in both sexes at 60 ppm, this study was uneventful. Histopathology examination included nasal tissues, and was uniformly negative. This satellite study was initially examined by Martz on 11/13/86, and was re-evaluated without an additional worksheet by Aldous on 1/15/15.

Chronic, dog ** †

****50046-061 117410** Stott, W.T., Stebbins, K. E., Haut, K. T., Quast, J. F., and Shabrang, S. N.; "Telone*II soil fumigant: One-year dietary toxicity study in beagle dogs", The Dow Chemical Co., Midland, Study ID M-003993-024, 7/22/92. Dogs were fed diets containing microencapsulated Telone*II at 0, 0.5, 2.5, or 15 mg/kg/day for 1 year. NOEL = 2.5 mg/kg/day [hematology profile typical of hypochromic, microcytic anemia: related to increased hematopoiesis in bone marrow and extramedullary hematopoiesis in spleen in both sexes]. Clinical signs in 2 high dose males of pale skin/mucous membranes apparently reflected the anemia. Body weights were depressed and relative liver weights were increased in both sexes at 15 mg/kg/day. The relatively low NOEL for signs of anemia constitutes a "**possible adverse effect**". **Acceptable**; Aldous, 11/15/93.

Oncogenicity, mouse ** † (adverse effects in inhalation guideline study only)

****50046-0029 060675** Stott, W. T., K. A. Johnson, L. L. Calhoun, S. K. Weiss, and L. E. Frauson, "Telone* II Soil Fumigant: 2-year inhalation chronic toxicity-oncogenicity study in mice," The Dow Chemical Company, Midland, MI, 7/13/87, Laboratory Study # M-003993-009. In the main oncogenicity study, fifty B6C3F1 mice/sex/group were dosed by whole-body inhalation for 6 hours/day, 5 days/week for a total of 510 exposure days in an oncogenicity study. [See DPR Record No. 036219 in Document No. 50046-0006 for the associated chronic study component (with 6-month and 1-yr sacrifices)]. Test article was 1,3-Dichloropropene, 92.1% (cis 49.5% and trans, 42.6%) plus 1,2-dichloropropane 0.7%, 1,3-dichloropropane 1.8%, 1-chlorohexane 1.1% and the remainder a mixture of isomers of chlorohexane, chlorohexene and dichloropropene, Lot TB831213-4. Nominal doses were 0, 5, 20, or 60 ppm (not corrected for purity). Exposure corresponds to 0, 22.7, 90.8, or 272.4 mg/m^3 nominal, uncorrected for purity of 92%, so content of the active ingredient was correspondingly lower (i.e., 20.9, 83.6, or 251 mg/m^3) adjusted for purity. NOEL = 5 ppm, based on urinary bladder mucosal hyperplasia in both sexes, and on hyperplasia and hypertrophy of the nasal respiratory epithelium (bilateral) in females. The nasal respiratory epithelium response was nearly universal in both sexes at 60 ppm. In addition, there were aggregations of lymphoid cells in the urinary bladder submucosa in

60 ppm females, hyperplasia of the non-glandular mucosa of the stomach in 60 ppm males, and degeneration of the olfactory epithelium of nasal passages in both sexes at 60 ppm. There was a statistically significant increase in incidence of the common tumor, bronchioalveolar adenoma, in 60 ppm males. This study is **acceptable**, with possible **adverse effects** (bronchioalveolar adenoma, major reactions in nasal respiratory and olfactory epithelia, and urinary bladder mucosal hyperplasia (J. Gee, 7/12/88). The study was re-examined by Aldous in conjunction with a risk assessment (1/14/15).

50046-0006 036219 Yano, B. L., L. L. Calhoun, W. T. Stott, K. A. Johnson, S. K. Weiss, and D. J. Schuetz, "Telone* II Soil Fumigant: 2-year inhalation chronic toxicity-oncogenicity study in mice – Interim report: 6- and 12-month exposures," The Dow Chemical Company, Midland, MI, 9/27/85 (for chronic phase), Laboratory Study # M-003993-009. In this chronic phase associated with an oncogenicity study (see DPR Record No. 060675), ten B6C3F1 mice/sex/group were dosed by whole-body inhalation for 6 hours/day, 5 days/week for either 6 or 12 months. Test article was 1,3-Dichloropropene, 92.1% (cis 49.5% and trans, 42.6%) plus 1,2-dichloropropane 0.7%, 1,3-dichloropropane 1.8%, 1-chlorohexane 1.1% and the remainder a mixture of isomers of chlorohexane, chlorohexene and dichloropropene, Lot TB831213-4. Nominal doses were 0, 5, 20, or 60 ppm (not corrected for purity). Exposure corresponds to 0, 22.7, 90.8, or 272.4 mg/m^3 nominal, uncorrected for purity of 92%, so content of the active ingredient was correspondingly lower (i.e., 20.9, 83.6, or 251 mg/m^3) adjusted for purity. NOEL for males = 5 ppm, based on bilateral hyperplasia and hypertrophy of the respiratory epithelium at 20 ppm at 1 year. NOEL for females = 20 ppm, based on hyperplasia and hypertrophy of the respiratory epithelium, epithelial hyperplasia of the urinary bladder in females, and subacute to chronic inflammation of the urinary bladder in females. Body weights were typically marginally reduced after 6-12 months (usually not statistically significant). This is a valid supplement to the oncogenicity study (original review was by F. Martz, 1/14/86), re-examined by Aldous in conjunction with a risk assessment (1/27/15).

50046-097 140561 Redmond, J. M., K. E. Stebbins and W.T. Stott, "Telone*II Soil Fumigant: Two-year dietary chronic toxicity/oncogenicity study in B6C3F1 Mice - Final Report", The Toxicology Research Laboratory, The Dow Chemical Co., Midland MI. Aug. 9, 1995. Study ID M-003993-032. Microencapsulated Telone*II, purity 95.8% 1,3-dichloropropene (50.7% cis/45.1% trans), was admixed with the diet at concentrations of 0, 2.5, 25, or 50 mg/kg/day and fed to 50 B6C3F1 mice/sex/group for 24 months. Additional mice (10/sex/group) were allocated for a 1-yr interim sacrifice. NOEL = 2.5 mg/kg/day (body weight decrements, both sexes). Upper dose levels achieved, but did not exceed, an MTD. The most definitively treatment-related effect was decreased hepatocyte size in 6/10 of the 50 mg/kg/day males at 1-yr sacrifice. There was a small increase in high dose females with stromal cell sarcomas, originating in the cervix or uterus, compared to concurrent controls (one control vs. four high dose females). In the 1996 DPR review, this was considered as a "possible adverse effect", and histopathological examinations of cervix or uterus slides of intermediate groups were requested, in addition to relevant historical control data. The requested data were submitted (Document No. 50046-103, Record # 151706). Stromal cell sarcoma incidence was 1, 0, 1, and 4 in controls through high dose groups. Historical control incidence of combined uterine or cervical stromal cell sarcomas ranged from 0 to 4, with 4/16 studies having either 3 or 4 such tumors out of 50 mice. **No adverse effect: data do not indicate a treatment effect on tumor

incidence. The study is now **acceptable as an oncogenicity study** (absence of blood chemistry precludes acceptance as a "combined" study). Aldous, 4/17/96, 3/10/97.

50046-072 126521 This 75-page interim report relates to Record # 140561 above. There are no essential data unique to this interim report. Aldous, 7/15/99.

Various mouse oncogenicity studies: non-guideline exposure protocols

50046-010 036553 "Toxicology and Carcinogenesis Studies of Telone II in F344/N Rats and B6C3F1 Mice." (NTP Technical Report Series No. 269, Frederick Cancer Research Center, 5/85). **This 1-liner summarizes results in mice**. Telone II (1,3-dichloropropene, 87.5% pure, with epichlorohydrin as stabilizer) was administered to 50/sex/group at 0, 50, or 100 mg/kg by oral gavage "3 times a week" for 104 weeks. Study is scientifically valid but unacceptable and not upgradeable because it was not designed to address FIFRA guidelines. Also, 25 control male mice died mid-study from myocarditis, compromising the usefulness of this group. **Urinary bladder** was a target organ, with primary non-neoplastic lesion of epithelial hyperplasia incidence (out of 50) in control through high dose males of 0, 9, and 18, and in females of 2, 15, and 19. Urinary bladder incidence of transitional cell carcinoma was 0, 0, and 2 for corresponding males, and 0, 8, and 21 in females. **Lungs** did not show pre-disposing non-neoplastic change, but there were increases in alveolar/bronchiolar tumor incidences as follows: control to high dose male adenoma incidences of 1, 11, and 9, respectively; and 0, 3, and 8 in females; corresponding carcinoma incidences were 0, 2, and 3 in males; and 2, 1, and 0 in females. **Forestomach** hyperplasia incidence was 0, 0, and 4 in males; and 1, 1, and 21 in females. Forestomach squamous cell papilloma incidences were 0, 2, and 3 in males; and 0, 1, and 2 in females. Forestomach squamous cell carcinoma was observed only in 2 high dose females. Study is not acceptable, due to compromised control male survival (above) and to technical problems in design. Above tumors and pre-disposing lesions are "possible adverse effects." Original review was by Martz, 1/17/86, with a revision of this summary without a new worksheet by Aldous, 1/30/15.

NOTE: Document No. 50046-010, Record No. 036554 refers to the same published article in JNCI 63 cited in Document No. 50046-007, Record Nos. 028361-028363. Multiple record numbers in Document No. 50046-007 represent 3 different dosing protocols presented in the publication, none of which approached guideline procedures. Aldous, 4/22/96.

50046-010 036554 "Carcinogenicity of Halogenated Olefinic and Aliphatic Hydrocarbons in Mice." (Van Duuren, B. L. et al., NYU Med Center, JNCI 63: 1433-1439, 1979) Ha:ICR Swiss strain; cis-1,3-dichloropropene (Chemical Samples Co., Columbus, OH), 122 mg/mouse or 41 mg/mouse by dermal application 3/week for about 77 weeks; initially reviewed as having caused no local or distant tumors. UNACCEPTABLE and not upgradeable. Reviewed: 6/3/85 by Apostolou, peer review 2/20 and 8/18/86 by Martz. Re-review as part of the risk assessment process noted that the incidence of lung tumors in both groups of treated mice was statistically significant by Fisher's Exact Test although not so noted in the publication table. The incidences were 30/100 for controls and 19/30 and 17/30 at low and high doses respectively. Study remains UNACCEPTABLE but with a possible adverse effect. (Gee, 5/31/90).

50046-007 028361 (same publication as 036554, above, refers to the repeated dermal cis-1,3-dichloropropene treatments).

010 036554 "Carcinogenicity of Halogenated Olefinic and Aliphatic Hydrocarbons in Mice." (NYU Med Center, JNCI 63: 1433-1439, 1979) Ha:ICR Swiss strain; cis-1,3-dichloropropene (Chemical Samples Co., Columbus, OH), 3 mg/mouse once weekly x 77 weeks by subcutaneous injection; examined injection site and liver only; fibrosarcoma at injection site, 6/30 vs. 0/30 vehicle control, probably due to irritation by physical-chemical properties of A.I. Otherwise, insufficient for assessment. UNACCEPTABLE and not upgradeable. (Apostolou, 6/3/85; Martz 2/20 and 8/18/86).

50046-007 028362 (same publication as 036554, above, relating to subcutaneous cis-1,3-dichloropropene treatment). Reviewed under this record number by Apostolou (see above).

010 036554 (suppl. to 028363) "Carcinogenicity of Halogenated Olefinic and Aliphatic Hydrocarbons in Mice." (NYU Med Center, JNCI 63: 1433-1439, 1979) Ha:ICR Swiss strain; cis-1,3-dichloropropene (Chemical Samples Co., Columbus, OH), 122 mg/mouse by a single dermal application, followed by promotion with 5 mg phorbol myristate acetate (PMA) dermally 3/week for about 77 weeks; no significant increase in tumors due to the a.i. [4/30 with dermal papillomas in cis-1,3-dichloropropene group: 6/90 in PMA positive control group: 0/100 in untreated mice, and evidently 0/30 in acetone (sham promoter) treatment group]. UNACCEPTABLE and not upgradeable. (Reviewed: 6/3/85 by Apostolou, peer review 2/20 and 8/18/86 by Martz).

50046-007 028363 (relates to investigation using single dermal application of cis-1,3-dichloropropene plus PMA promotion in Record No. 036554, above: originally reviewed under this record number by Apostolou).

GENOTOXICITY

Bacterial reverse mutation assay ** † (positive studies may not represent modern Telone II a.i.)

The series of genotoxicity studies submitted for 1,3-dichloropropene contains many positive studies and many negative studies. The positive studies tend represent older formulations of technical 1,3-dichloropropene (typically pre-1980), whereas negative studies are typically several years more recent. Usually Ames test studies which indicate positive bacterial mutagenicity responses found base-pair substitution mutants rather than frameshift alterations (i.e. tests show increased revertants in TA 1535 and TA 100 strains, compared to little or no response in strains TA 98, TA 1537, or TA 1538). Usually the presence or absence of S-9 has little to do with mutagenicity. Providing human liver physiological levels of glutathione (GSH) typically greatly suppresses mutagenicity. Many supplementary studies relating to oncogenicity and mutagenicity are found below beginning with the section on "Mechanistic Studies," below. Aldous, 6/22/15.

50046-016, **004282** & 004293 "Mutagenicity of 1,3-Dichloropropene using Ames Testing." (Schering AG, summary report 9/82) Formulated mixtures containing 1,3-dichloropropene in addition to various other constituents, were tested for mutagenic activity in the Ames Salmonella Test. Results were conflicting and insufficient for independent assessment. UNACCEPTABLE but upgradeable upon submission of complete report(s). Summary contains statement that positive effects were seen with TA1535 and TA100 but no data. Report contains a statement that the methyl isothiocyanate in the sample tested caused cytotoxicity before the mutagenic effect was detectable. No data. (Reviewed: 6/3/85 by Apostolou, peer review 8/18/86 by Martz and 7/18/88 by Gee).

Note by Aldous (4/20/15) relating to the above Schering AG study: the test article in this study was designated as "D-D," a mixture of "C_3 hydrocarbons including dichloropropenes, dichloropropene and related chlorinated hydrocarbons." This mixture constituted 80% of a formulation called Vorlex II, which is not currently registered in California. This test article differs appreciably from modern Telone II. The NTP report of rat and mouse oncogenicity studies reported in 50046-0010, Record No. 036552 and 036223, describes D-D as a synonym of *cis, trans*-1,3-dichloropropene, as found in Telone and Vorlex Soil Fumigant products. The latter record specifies components of the technical test article as including 89% *cis, trans*-1,3-dichloropropene, 1.5% of a trichloropropene isomer, 2.5% 1,2-dichloropropane, and most importantly, 1.0% epichlorohydrin. Technical Telone II from a more recent assay (Sept. 1999) was comprised of 97.5% *cis, trans*-1,3-dichloropropene, with lesser amounts of trichloropropene isomers and of 1,2-dichloropropane, and no epichlorohydrin. It is unclear to what extent contaminants in older technical 1,3-dichloropropene may have affected the mutagenicity tests.

50046-0010 036556 "Mutagenicity of 1,3-Dichloropropene in Bacteria Test System." (Nomura Sogo Res. Inst., 12/78) E. coli strain B/r, Wp 2, Try$^-$; 49.8%-cis and 46.3%-trans 1,3-dichloropropene, 5000, 2500, 1000, 500, 250, 100, 25, or 0 µg/plate, ±S9. Unacceptable and not upgradeable due to design deficiencies. No mutagenic effects were reported. (Gee, 2/24/86).

50046-010 036558 "Mutagenicity of 1,3-Dichloropropene in Bacteria Test System." (Nomura Sogo Res. Inst., 12/78) Five Salmonella strains for plate assay; 49.8%-cis and 46.3%-trans 1,3-dichloropropene, 0-5000 µg/plate ± S9; G46 for host-mediated assay in ICR mice at 30 or 60 mg/kg x 3 times/3 hours. UNACCEPTABLE and not upgradeable: single plates. Significant **Positive response** in several strains indicative of base-pair substitution; negative in host-mediated assay. (Gee, 2/24/86).

Note (by Aldous, 4/21/15): there were > 10-fold increases in revertant colonies, particularly for *Salmonella typhimurium* strains TA 1535 and TA 100, with and without S-9. The report does not state whether this technical product contained epichlorohydrin. This study was of the same time period as the NTP studies in rats and mice, which tested an old formulation technical 1,3-dichloropropene containing 1% epichlorohydrin. See Document No. 50046-010, Record Nos. 036552 and 036553, "Toxicology and Carcinogenesis Studies of Telone II in F344/N Rats and B6C3F1 Mice."

(No DPR Document or Record Number) De Lorenzo, F., S. Degl'Innocenti, A. Ruocco, L. Silengo, and R. Cortese, "Mutagenicity of pesticides containing 1,3-dichloropropene," Cancer Research 37:1915-1917 (June 1977). Investigators reported that an old technical formulation of Telone elicited several-fold increases in revertants over controls in point mutation-associated

Salmonella strains TA 1535 and TA 100, and also in TA 1978 (described as similar to TA 1538, but with a normal DNA-excision repair system). Investigators also tested individually the *cis*- and *trans*- isomers of 1,3-dichloropropene, reporting roughly 50-fold increases in mutant colonies with and without S9 in TA 1535, roughly 20-fold increases with and without S9 with TA 100, and about 4-fold increases with TA 1978: all of these maximum responses were in the range of 50 to 100 µg/plate. In addition to the major Telone ingredients, investigators also evaluated 2,3-dichloro-1-propene, which elicited responses similar to the Telone isomers, and 1,2-dichloropropane, which was about 1000-fold less potent than the Telone isomers. This was a 3-page report, with no evident quality assurance protocols and no individual data to validate reported results. As such, there is no DPR worksheet. One-liner is by C. Aldous, 4/22/15.

Mutagenicity: *In vitro* mammalian cell assay **

Summary: Typically mammalian cell studies have been negative. Some older positive bacterial studies have been reported. There are no recent, standard Ames tests employing modern formulation Telone® II (which does not have mutagenic epichlorohydrin as stabilizer). This is important, because older studies (such as Record No. 036558) which were positive did contain epichlorohydrin. One problem is the volatility of the test material and care must be taken to control samples for this property. From the text of the study with CHO, the flasks were tightly capped and loss of test material should not have been a factor. Overall, as of 9/10/99, there is considered to be a possible genotoxic effect in bacteria unless there are more recent studies using the current test article. Gee, 7/18/88 and 9/10/99.

** 019 042945 "The Evaluation of Telone II Soil Fumigant in the CHO Cell/HGPRT Forward Mutation Assay." (Dow, 2/27/86) CHO/HGPRT assay; Telone II (48.9% cis and 43.2% trans 1,3-dichloropropene); 250, 200, 150, 100, 50, or 0 mM without S9 (3 trials) and 200, 150, 125, 100, 50, or 0 mM with S9 (1 trial). Report complete and study ACCEPTABLE. NO evidence of mutagenicity. (Gee, 7/24/86).

Mutagenicity: *In vivo* cytogenetics **

** 010 036560 "Evaluation of Telone II Soil Fumigant in the Mouse Bone Marrow Micronucleus Test." (Dow, 5/85) Telone II (49.5%-cis and 42.6%-trans 1,3-dichloropropene), 380, 115, 38, or 0 mg/kg by oral gavage in CD-1 mice, 5/sex/group, 24 or 48 hour sacrifice. Reviewed 2/25/86 as incomplete but upgradeable with justification of the use of only two sacrifice times. This has been submitted as Record #55630 in 50046-025, based on excretion of 93% within 48 hours. The study is now reviewed as ACCEPTABLE. NO increase in micronucleated PCE's reported. (Gee, 2/25/86 and 4/16/87).

**50046-115 162466 Gollapudi, B. B., F. S. Cieszlak, and S. J. Lick, "Telone* II soil fumigant (cis/trans 1,3-dichloropropene): inhalation dominant lethal mutagenicity study in the CD (Sprague-Dawley derived) rat", The Dow Chemical Co., Midland, 5/29/97. Laboratory Project Study ID 960035. Thirty male Crl:CD®(SD) rats per treatment group were dosed by inhalation for 6 hr/day, 7 days/wk, 10 weeks duration at levels of 0, 10, 60, and 150 ppm. Negative pair-fed controls (matched to food consumption of high dose rats) and positive controls (single oral dose of cyclophosphamide given 48 hr prior to first mating period) were not housed in inhalation chambers. Each of these male treatment groups consisted of 30 rats. There were two consecutive mating periods of 1 week each during weeks 11 and 12 (1 male/2 females). On day

13 after the end of respective mating periods, females were euthanized. Corpora lutea were counted, and uteri were examined for numbers of live implants and resorption sites. Uteri of apparently non-pregnant females were stained with sodium sulfide and examined for possible early resorptions. The NOEL for "subacute" change = 10 ppm (decreased food consumption and decreased body weight, particularly during the first week of treatment). There was no evidence of a dominant lethal effect (no increase in resorptions). **Acceptable, with no adverse effects**. Aldous, 8/18/99.

Mutagenicity: DNA Damage, or uncommon study designs ** †

Summary: Different tests measure different endpoints so no one conclusion can be reached. A possible adverse genotoxic effect is noted. As was the problem with point mutation studies above, older investigations employing mutagenic epichlorohydrin as stabilizer may not be relevant to evaluation of modern formulation Telone® II. Gee, 7/18/88 and 9/10/99.

** 010 036559 "Evaluation of Telone II in the Rat Hepatocyte Unscheduled DNA Synthesis Assay." (Dow, 4/85) UDS in rat hepatocytes; Telone II (49.5% cis and 42.6% trans-1,3-dichloropropene) $1x10^{-7}$ to $3x10^{-3}$ M concentration (solubility limit), plus control. Report complete and study ACCEPTABLE. NO evidence of UDS even when cytotoxicity was noted. (Gee, 2/24/86).

010 036557 "Mutagenicity Test on 1,3-Dichloropropene in Bacteria Test System." (Nomura Sogo Res. Inst., 12/78) Bacillus subtilis rec assay, strains H17 and M45; 49.8% cis- and 46.3% trans-1,3-dichloropropene, 1250, 500, 125, 50, or 0 mg/well without activation. UNACCEPTABLE and not upgradeable due to design deficiencies. **Slight growth differences at highest level.** Reviewed 2/24/86 by Gee. See also 1-liner for study 50046-010 036558, above, noting the likelihood epichlorohydrin in technical product of that time period.

50046-119 162470 Stott, W. T., T. J. Miller, and A. K. Wardynski, "1,3-Dichloropropene: *in vitro* DNA binding", The Dow Chemical Co., Midland, 12/12/97. Laboratory Project Study ID 970180. The study evaluated adduct formation when ^{14}C-labeled test material was incubated with calf thymus DNA solution with appropriate co-factors. Functional positive controls were ^{14}C-methyl iodide (without S9) and ^{14}C-1,2-dichloroethane (with S9). ^{14}C-1,3-dichloropropene did not elicit binding with or without S9. Useful ancillary data. The study does not address FIFRA data requirements, and was not performed under QA oversight. Aldous, 9/23/99.

50046-111 161850 Exact duplicate of 50046-119 162470, above.

Note: The reregistration standard of 1986 noted requirements for in vitro/in vivo primary hepatocyte UDS testing both in vitro and in vivo exposure - species not specified. Record # 036559 is not cited. (Gee, 5/23/89).

No record number "Chemical Mutagenesis Testing in Drosophila. III. Results of 48 Coded Compounds Tested for the National Toxicology Program." (Valencia, R., Mason, J. M., Woodruff, R. C., and Zimmering, S., Environmental Mutagenesis 7: 325 - 348 (1985)) 1,3-Dichloropropene technical, 95.5% was tested with male Canton-S wild-type stock by feeding at 5,570 ppm for 72 hours from soaked filter paper. The males were mated to Basc females for 3, 2 and 2 days. No more than 40 females per parental male were mated from each brood. A total of

6584 tests were performed. The percent lethals were 0.12 for control broods and 0.30 for treated broods - **considered positive by the authors.** The translocation test was negative. No worksheet. [Review was done in connection with the risk assessment.] (Gee, 5/31/90).

50046-120 162475 Gollapudi, B. B. and Cieszlak, F. S., "Telone® II Soil Fumigant: Evaluation in an *in vivo* assay for gene mutagens using transgenic Big Blue Mice," The Dow Chemical Co., Midland, 2/10/97. Laboratory Study # K-006409-017. Male Big Blue B6C3F1 mice, 5/group, were dosed by inhalation at 0, 10, 60, or 150 ppm of Telone II® Soil Fumigant, 96% purity, for 10 exposures (5/week over 2 weeks), at 6 hours/day. After an additional 17-day expression period, mice were killed. Each cell of the test mouse had about 40 copies of a shuttle vector carrying the *lacI* gene, the *lacI* promotor, the *lacI* operator, and the *αlacIZ* reporter gene. Mouse tissues (lung and liver) were homogenized, and the DNA was collected, digested, and packaged into phage particles using a proprietary system. The packaged DNA was added to plates containing the *E. coli* host bacteria. Following incubation, investigators counted the numbers of blue plaques compared to the total numbers of plaques as an index of mutations of the *lacI* gene. Blue plaques occur when a defective repressor protein allows transcription of the reporter gene, the product of which cleaves a chromogenic substrate (X-gal) in the medium. Only controls and 150 ppm mice were evaluated. Results showed no increases in mutations in lung or liver. Functional positive control tissues evidently derived from a single mouse, which was treated with five daily doses of 15 mg/kg/day diethylnitrosamine in water 54 weeks before sacrifice. NOEL for general toxicity = 60 ppm: all 150 ppm mice showed "decreased activity" during days 3-5 and days 8-12. There were no clinical signs observed at any other dose level. Body weights were not affected. Study is **not acceptable but is upgradeable** (DPR review notes concern about positive control). Aldous, 9/23/99, and again by Aldous on Jan. 8, 2015 (for minor additions to 1-liner to highlight subacute effects).

50046-111 161855 Exact duplicate of 50046-120 162475, above.

REPRODUCTIVE TOXICITY, RAT **

**50046- 030 060676 Breslin, W. J., H. D. Kirk, C. M. Streeter, J. F. Quast, and J. R. Szabo, "Telone II Soil Fumigant: Two-Generation Inhalation Reproduction Study in Fischer 344 Rats." (Dow Chemical, 7/13/87, M-003993-015). Thirty rats/sex/group received 1,3-dichloropropene, 91.2%, lot #TB831213-4, by inhalation at 0, 10, 30 or 90 ppm for 5 days/week during pre-mating periods, and for 7 days/week during mating, gestation, and lactation periods. This treatment schedule was continuous except that gravid females were not dosed from presumed gestation day 20 until lactation day 4. There were two generations with two littering periods each. Pups were not directly exposed, but were separated from dams for the 6-hour maternal exposures. Parental NOEL = 30 ppm, based on decreased adult body weights (commonly statistically significant in males and occasionally significant in females), and nasal histopathology (slight hyperplasia of the respiratory epithelium in the majority of adults, and degeneration of the olfactory epithelium in many adults at 90 ppm). Reproduction NOEL $\geq$ 90 ppm (no adverse effect on reproduction parameters). Acceptable. (Gee, 7/13/88, with a re-examination that did not make major changes to the 1-liner, with no new worksheet, by Aldous on Jan. 8, 2015).

010 036555 "D-D: A 10 Week Inhalation Study of Mating Behavior in Male and Female Rats." (Shell (UK), 4/80) Wistar strain; technical D-D ("epichlorohydrin free"), 53.7% 1,3-

dichloropropene, remaining constituents mainly chlorinated isomers/analogs; 96, 32, 14, or 0 ppm for 6 hours/day x 5 days/week; treated males mated with naive females after 2, 4, 7, and 10 weeks exposure; treated females mated with naive males after 10 weeks exposure; hematology, serum chemistry, urinalysis, and histopathology on satellite animals; 30 males and 24 females per group with 20 and 15 respectively for reproduction performance and the remainder for hematology, etc. UNACCEPTABLE and not upgradeable: only 1 generation and inadequate group sizes. Otherwise, appears to be a well conducted and documented study with scientifically valid results. NO reproductive effects. Liver and kidney weight elevation at 96 ppm, reversible upon withdrawal, except female kidney values. (Martz, 2/20/86) (Report is apparently identical to Document No. 50046-013, Record No. 932846).

DEVELOPMENTAL TOXICITY

Rat Developmental Toxicity**

50046-0010 036561 John, J. A., P. M. Kloes, L. L. Calhoun, and J. T. Young, "Telone* II: Inhalation teratology study in Fischer 344 rats and New Zealand White rabbits," The Dow Chemical Company, Midland, MI, 10/31/83. [Rat and Rabbit data were presented together in this report: the present DPR review is confined to the **rat.] Thirty mated female F344 rats were dosed with Telone II (1,3-dichloropropene; 90.1% pure: 47.7% *trans* and 42.4% *cis* isomers) by whole-body inhalation at 0, 20, 60, or 120 ppm for 6 hrs/day on gestation days 6-15 in a developmental toxicity study. A review by Martz (2/21/86) found study to be **acceptable, with no adverse effects**, setting the maternal effects NOEL at less than 20 ppm, based on reduced maternal body weight gain associated with reduced food consumption at all dose levels. Martz attributed a modest increase in delayed ossification of vertebral centers at 120 ppm to treatment, justifiably attributing this to maternal toxicity (body weight loss). The noted change in ossification delay in vertebral centra (affecting 6/27 control and 12/24 high dose litters) was not statistically significant. This places the NOEL for developmental response at 60 ppm. This study was re-examined by Aldous in 2015 to re-examine the basis of the NOEL. Re-examination confirms that there is no maternal NOEL (NOEL < 20 ppm): food consumption was reduced with linear dose-response throughout the treatment period in all treatment groups. Maternal body weight gain underwent a modest but statistically significant decrement at onset of dosing (gestation days 6-8) at 20 ppm. Maternal body weight gain was significantly reduced in 60 and 120 ppm dams during gestation days 6-11. Aldous, Jan. 8, 2015. (See pilot study, rat and rabbit, below.)

50046-0223 283293 Kloes, P. M., L. L. Calhoun, J. T. Young, and J. A. John, "Telone II: Inhalation teratology probe study in Fischer 344 rats and New Zealand White rabbits," The Dow Chemical Company, Midland, MI, March 30, 1983. Mated female rats and rabbits were dosed by whole-body inhalation for 6 hr/day at 0, 50, 150, or 300 ppm of Telone II (47.7% cis, 42.2% trans, with impurities including 1.8 % epichlorohydrin) on gestation days 6-15 (rat) or 6-18 (rabbit). Scheduled sacrifices were on gestation days 16 (rats) or 19 (rabbits). RAT PHASE: There were 8, 7, 8, and 8 dams on study (control through 300 ppm groups), yielding 8, 7, 6, and 7 dams with litters at sacrifice. One high dose dam was found dead on gestation day 14: all others survived. Two of the eight 150 ppm dams had pregnancies that were only evident upon sodium sulfide staining of the uteri: this is unlikely to have been treatment-related, since the 300 ppm group had recognizable fetuses in all litters. Litters/[litters with resorption(s)] were 3/8, 3/7,

3/6, and 7/7 in control through high dose groups (statistically increased and likely dose-related at 300 ppm). Censoring the two mid-dose cases of sulfide-staining-only pregnancy detection, incidences of litters totally resorbed were 0/8, 0/7, 1/6, and 3/7, respectively. Treatment effect may be supposed for 300 ppm rats, whereas the single case at 150 ppm was equivocal [note that in the definitive developmental toxicity study (DPR Record No. 036561), 0/24 pregnant dams had total litter losses at the highest dose of 120 ppm]. Rat body weight gain decrements were evident at all dose levels, particularly during the first 2 treatment days. Body weight gains between gestation days 6 and 8 were 3.5, -4.5, -14.3, and -21.4 g in respective groups (statistically significant in all treated groups). Subsequent body weight gains were normal for 50 ppm dams, but body weight decrements continued for 150 ppm and especially for 300 ppm dams, with net body weight gains of 26, 13, -9, and -51 g in respective groups. Food consumption during the first 3 treatment days was unaffected at 50 ppm, but markedly reduced at 150 and 300 ppm (13, 13, 9, and 3 g/rat/day). Reduced food consumption was evident through gestation day twelve at 150 ppm and throughout the study at 300 ppm. There were no tabulated clinical signs data in this study, but investigators observed that "rats exposed to 300 ppm of TELONE II were observed to have urine and fecal staining as early as day 8 of gestation. Nasal exudate and red crusty material around the eyes were observed in this group intermittently until necropsy on day 16 of gestation." No clinical signs were evident in 50 or 150 ppm dams. RABBIT PHASE: There were 7 mated does/group on study, yielding 6, 7, 6, and 0 does with litters at sacrifice (one control was not pregnant, one 150 doe died of a bacterial pneumonia evidently unrelated to treatment, and all six pregnant 300 ppm does died). Investigators found no clinical changes in 50 or 150 ppm does, however "Six of 7 rabbits exposed to 300 ppm of TELONE II, however, showed signs of toxicity such as rear limb ataxia, decreased or absence of righting reflex and flaccid hind limb muscles. These animals were either sacrificed moribund or found dead within 24 hours of onset of these signs. Onset of these signs occurred between gestation days 14 and 19." Body weights were not affected at 50 ppm, but were decreased at 150 ppm only (body weight gain during treatment phase was 18, 17, and -202 g for control through 150 ppm respectively: with no 300 ppm females surviving to termination). There were no treatment effects at gestation day 19 examination of fetuses of 50 or 150 ppm does, with no pregnant surviving does at 300 ppm. Useful supplementary data (pilot study), Aldous, 3/13/15. (Note: Record No. 283292 is an electronic submission of the present record.).

Rabbit Developmental Toxicity**

**50046-0010 036562 John, J. A., P. M. Kloes, L. L. Calhoun, and J. T. Young, "Telone* II: Inhalation teratology study in Fischer 344 rats and New Zealand White rabbits," The Dow Chemical Company, Midland, MI, 10/31/83. [Rat and Rabbit data were presented together in this report: the present DPR review is confined to the rabbit.] Twenty-five to 31 inseminated New Zealand White rabbits were dosed with Telone II (1,3-dichloropropene; 90.1% pure: 47.7% *trans* and 42.4% *cis* isomers) by whole-body inhalation at 0, 20, 60, or 120 ppm for 6 hrs/day on gestation days 6-18 in a developmental toxicity study. There were 24, 18, 17, and 21 does with viable litters at C-section, considered to be sufficient for evaluation. There was no evidence of reduced maternal viability or of altered developmental outcomes. The initial review by Martz (2/21/86) requested historical control data on soft tissue alterations as a condition of acceptance. Those data were provided in Document 50046-025, Record No. 50619. Upon receipt of the new information, Martz determined that this study is acceptable, with no adverse effects (3/26/87). Martz determined that the NOEL was 20 ppm for reduced maternal weight gain. This study was re-examined by Aldous in 2015 to re-examine the basis of the NOEL. The new examination

noted some deficiencies in this study compared to modern developmental toxicity studies, but the study remains acceptable. Examination of the individual body weight gain data shows that the evidence of decrements in maternal body weight gain during the first 3 days of dosing is equivocal. Study authors had concluded that the small body weight decrements during gestation days 6-8 at 60-120 ppm were plausibly treatment-related. The Aldous re-examination does not change the NOEL, but acknowledges that apparent body weight changes were small and lacking in dose-response. Aldous, Jan. 8, 2015. (See pilot study, rat and rabbit, above.)

NEUROTOXICITY (Hen studies are not currently required)

Acute neurotoxicity, rat

90-day neurotoxicity, rat

Developmental neurotoxicity, rat

Delayed neurotoxicity, hen

IMMUNOTOXICITY

ENDOCRINE DISRUPTOR STUDIES

APPENDIX

The Appendix includes published and unpublished studies. Often the published studies do not contain individual data, and typically lack quality assurance validation, and thus are not amenable to DPR review worksheets. Entries without DPR Document numbers and Record numbers were typically not submitted by registrants. Studies in this section were not undertaken to fill FIFRA data requirements, and do not appear to indicate unique evidence of toxic hazards. Collectively, these studies greatly enhance the understanding of mechanisms of toxicity beyond the scope of FIFRA-mandated protocols.

Mechanistic studies examined by DPR Data Review Group

50046-0026 058182 Dietz, F. K., E. A. Hermann, P. E. Kastl, D. A. Dittenber, and J. C. Ramsey, "1,3-dichloropropene: pharmacokinetics, effect on tissue non-protein sulfhydryls, and macromolecular binding in Fischer-344 rats and B6C3F1 mice following oral administration," The Dow Chemical Company, Midland, MI, March, 1985. This is a brief report of a relatively old study, without great detail, but useful primarily in addressing depletion of non-protein

sulfhydryls (below). Most of the ^{14}C-1,3-dichloropropene (1,3-D) in this study was uniformly labeled, with isotope ratio of 62% *cis*, 38% *trans* (a single urinary elution profile from rats treated with purified labeled *cis* 1,3-D was similar to those derived from the labeled cis/trans mixture). Only male rats and mice were used. Percent of administered label recovered in 48-hr experiments following 1 mg/kg doses was: (rat) 51% in urine, 20% in feces, 18% in exhaled CO_2, and 6% in carcass; (mouse) 79% in urine, 16% in feces, 14% in exhaled CO_2, and 2% in carcass. Tissue distribution (µg equivalents) 48 hours following 1 mg/kg dosing was: (rats) about 0.2 to 0.3µg equiv. for non-glandular stomach, glandular stomach, liver, kidney, and bladder; (mice) 0.23 µg equiv. for non-glandular stomach, about 0.1 µg equiv. for bladder, liver, and kidney, and only 0.04 µg equiv. for glandular stomach. Estimated elimination $T_{1/2}$ for rats and mice was consistently about 5-6 hrs, regardless of dose (1 and 50 mg/kg tested for rats, and 1 and 100 mg/kg for mice). Chromatographic analysis of rat and mouse urine samples found 2 prominent urinary peaks for each species: one peak was shown by MS to be N-acetyl-S-(3-chloroprop-2-enyl) cysteine derivatives, and the other peak was presumed to represent derived sulfoxide and/or sulfone products. Low dose (1 mg/kg) rats produced comparable amounts of the two products. Higher dose rats (30 to 50 mg/kg) produced roughly 3-fold more of the N-acetyl-S-(3-chloroprop-2-enyl) cysteine than sulfoxide and/or sulfone products. High dose mice (100 mg/kg) produced slightly more of the sulfoxide and/or sulfone products compared to N-acetyl-S-(3-chloroprop-2-enyl) cysteine. As had been previously reported in an abstract (see DPR Document No. 50046-0015, Record No. 932856), there was substantial difference in depletion of non-protein sulfhydryls (NPS) between different tissues. Non-glandular stomach showed the greatest depletion (evidently meaningfully reduced at 5 mg/kg in both species), and with marked depletion at 50 to 100 mg/kg in both species. In both species, NPS content of kidney was unaffected, the bladder scarcely affected, and glandular stomach and liver showed depletion to an intermediate degree. A time-course evaluation of NPS contents of these tissues in 100 mg/kg mice found complete recovery of NPS content in non-glandular and glandular stomach over the time frame of 4 to 8 or 12 hours after dosing. Liver, although never as profoundly depleted of NPS as the non-glandular or glandular stomach, required between 8 and12 hours to show recovery. Particularly the evidence that high doses of 1,3-D depletes NPS such as glutathione in non-glandular stomach may be relevant evidence for a threshold for tumors in this organ. Aldous, 1/30/15.

50046-120 162472 Lawlor, T. E., "Evaluation of 1,3-dichloropropene for mutagenic potential in *Salmonella* in the presence of mouse lung homogenate (S9)", Corning Hazleton Inc. (CHV), 11/26/96. CHV Study ID No. 17037-0-401. *Salmonella typhimurium* TA 100 was the only strain employed in this supplemental mutagenicity study, which evaluated the effects of S9 from mouse lung homogenate and of supplementary GSH in Ames-style reverse mutation plate assays following a 20-minute pre-incubation period in sealed tubes. S9 preparations were prepared from B6C3F1 mice: either controls, or exposed to 1,3-dichloropropene by inhalation (63 ppm, 5 days/wk, 2.5 wks). Preliminary tests with mouse lung S9 preparations with positive control substances benzo(a)pyrene and 2-aminoanthracene found that only the latter increased revertant incidence with mouse lung S9 mix. In studies with mouse lung S9 (whether derived from controls or from 1,3-dichloropropene-treated mice, in either case with or without GSH) there was no increase in revertants due to 1,3-dichloropropene treatment over the survivable range of 75 to 300 µg/plate. Studies without S9 similarly showed no treatment effect throughout the meaningful range. Cytotoxicity was evident between 300 and 600 µg/plate with S9. Studies

without S9 found no cytotoxicity at 300 μg/plate, but severe cytotoxicity at 450 μg/plate. Results were thus negative under conditions of study (single strain, single trial, three reps/dose level), suggesting that mouse lung microsomal enzymes did not elicit previously-indicated responses by metabolizing 1,3-dichloropropene to a mutagenic intermediate. Useful supplementary data. Aldous, 9/21/99.

50046-111 161852 Exact duplicate of 50046-120 162472, above.

50046-119 162471 Stott, W. T., B. B. Gollapudi, C. M. Clements, V. A. Linscombe, D. A. Dittenber, S. J. Lick, and K. A. Johnson, "1,3-Dichloropropene: mechanism of tumorigenicity studies in male B6C3F1 mice and Fischer 344 rats", The Dow Chemical Co., Midland, 12/12/97. Laboratory Project Study ID # 971121. Hepatocellular tumors were noted in male F344 rats administered microencapsulated Telone in the diet (Record No. 140562), and benign lung bronchioalveolar adenomas were previously found in male B6C3F1 mice in an inhalation study (Record No. 060675). The present study sought to determine whether Telone is mutagenic or otherwise likely to have elicited such tumors by any of several means evaluated, including: (1) cell proliferation studies: evaluated by analysis of BrdU uptake in target tissues, with nuclear labeling visualized by immunohistochemical methods, (2) apoptosis: evaluated by using antibodies to the exposed 3'-OH ends of DNA fragments (which are characteristic of apoptosis), followed by antibody binding, then by addition of a chromophore to the antibody constituents, allowing quantitation by light microscopy (3) Glutathione (GSH) was evaluated from rat liver and mouse lung homogenates by a clinical chemistry analyzer (in addition to sacrifices of animals exposed continuously up to the time of sacrifice, some rats and mice were killed about 24 hr after the last exposure to assess "rebound" recovery of GSH in tissues), (4) DNA adduct formation was evaluated by a ^{32}P-post-labeling assay, in which DNA was isolated, digested to release 3'-mononucleotides, enriched in certain fractions expected to contain adducts: these fractions were then labeled using T4 polynucleotide kinase and [γ-^{32}P]ATP, and finally treated with nuclease P1 to create labeled 5'-mononucleotides. Labeled adducts were then separated by 2-dimensional TLC. Chromatograms were compared to those produced by action of a known mutagen (propylene oxide) on DNA samples *in vitro*. NOEL's in rats and mice were 12.5 mg/kg/day and 10 ppm, respectively, based on reductions in GSH levels. NOEL's for outcomes more commonly considered to reflect toxicity, such as modest body weight decrements in both species and increased circulating ALT and AST in rats as indicators of liver responses, were 25 mg/kg/day and 60 ppm, respectively. Evidence of strong dose-response for depletion of GSH and increased turnover of GSH suggest that high dose effects may relate to depletion of natural detoxification capacity, indicating that high dose responses, including tumors, may have little or no relevance to chronic exposure at lower levels. None of the mechanistic studies identified treatment effects on cell proliferation, apoptosis, or DNA adduct formation. The authors concluded that the tumors were caused by a non-genotoxic mode of action *in vivo*. Often, small sample sizes and large inter-animal variability limited the level of confidence in these "negative" results. Aldous, 9/23/99.

50046-111 161851 Exact duplicate of 50046-119 162471, above.

Schneider, M., Quistad, G. B., and Casida, J. E. 1998. "1,3-Dichloropropene epoxides: intermediates in bioactivation of the promutagen 1,3-dichloropropene," *Chem Res Toxicol*

11:1137-1144. The 92% cis/trans (1:1) 1,3-dichloropropene (1,3-D) used in this study contained 0.2% each of cis and trans epoxides, which could be quantitatively removed by DMSO at room temperature (i.e. degraded by this solvent without the need for added cofactors). The metabolic oxidation of epoxide-free 1,3-D [cis/trans (1:1)] to form respective epoxides was demonstrated in Swiss-Webster mice (700 mg/kg by intraperitoneal injection): liver extracts of these mice yielded several times more cis- epoxide than trans- epoxide. Similarly, *in vitro* mouse liver microsomal preparations of epoxide-free 1,3-D in the presence of NADPH also yielded preferentially cis epoxide. When GSH in addition to NADPH were added to the same *in vitro* system, there were markedly reduced cis and trans epoxide levels, and respective parent 1,3-D levels in liver extracts were reduced to about 30% of untreated controls. Potential metabolites sought but not found in this assessment were 2-chloroacrolein and cis- and trans- 3-chloroacrolein. *In vivo* treatment of mice with either purified cis- or trans-1,3-D yielded only the corresponding cis- or trans- epoxide in extracted liver. Liver levels of cis- or trans- 1,3-D and corresponding epoxides were assessed following a single ip dose of 1:1 cis:trans 1,3-D. Of parent compounds, the trans-1,3-D levels predominated (roughly 2-fold) over cis-1,3D throughout the 150 minute sampling period. In contrast, cis- 1,3-epoxide predominated over the trans-epoxide in liver extracts by about 2-fold during that period. Cis/trans 1,3-D epoxides dissolved in DMSO yielded a virtually quantitative product of 2-chloroacrolein within 280 minutes at 22°C, with 2,3-dichloropropanal as an intermediate product. Cis/trans 1,3-D epoxides decomposition in pH 7.4 buffer yielded predominantly 3-chloro-2-hydroxypropanal (including its dimer). Whereas both epoxides degraded spontaneously in pH 7.4 buffer, addition of GSH roughly doubled degradation rate, and GSH plus glutathione S-transferase (GST) increased the latter rate about 60-fold. In contrast, 2-chloroacrolein (not detected in this study under physiological conditions) degraded only slowly in buffer alone, but decomposed too rapidly to quantify upon addition of GSH, with or without GST. Rat liver microsomal epoxide hydrolase and soluble epoxide hydrolase activities, when expressed in insect cells, were examined for inhibition by cis/trans-1,3-D epoxides of activity toward tritiated cis-stilbene oxide. When 1 mM solutions of cis/trans-1,3-D epoxides were incubated for various time periods prior to addition of cis-stilbene oxide in the presence of rat liver **microsomal** epoxide hydrolase, there was about 60% inhibition of cis-stilbene oxide hydrolase activity at time 0, with a decline to zero inhibition after 10 minutes. When 1 mM solutions of cis/trans-1,3-D epoxides were incubated for various time periods prior to addition of labeled trans-stilbene oxide in the presence of rat liver **soluble** epoxide hydrolase, the trans-stilbene oxide hydrolase activity inhibition peaked in 5 minutes at 50% inhibition, with about 35% inhibition remaining at the final sampling time of 20 minutes. **Microsomal** epoxide hydrolase is associated with many xenobiotic responses. Mutagenicity by standard plate incorporation method in *S. typhimurium* TA 100, a system previously shown respond to technical Telone, found mutagenicity of 1,3-D (cis) to be at least 4 orders of magnitude less potent than either of the 1,3-D epoxides (trans 1,3-D was not tested here). A very small positive value reported for 1,3-D (cis) **with** S9 activation was considered to represent a treatment effect by investigators. Both epoxides elicited revertant responses, with cis-epoxide about 2x more potent than trans-epoxide. As observed in other studies, S9 had no effect on mutagenicity of either of the epoxides. 2-Chloroacrolein was about 4x more potent than cis-1,3D epoxide. In contrast, 3-chloroacrolein (also not shown here to be a metabolite of 1,3-D under physiological conditions) was a very weak direct mutagen, about the same potency as cis-1,3-D. Varying GSH from 0 to 5 mM in TA 100 plate incorporation tests employing cis- and trans-epoxides (without GST) yielded a log-linear decrease in revertants over the entire range tested, although the cis-epoxide

retained some mutagenicity even at the highest GSH level. Under these conditions, 2-chloroacrolein showed roughly constant mutagenicity between 0 and 0.25 mM GSH, with mutagenicity diminishing to about 25% of maximum levels at 5 mM GSH. In the latter test series, with cis- and trans-epoxides and with 2-chloroacrolein, addition of glutathione S-transferase either had no effect or decreased mutagenicity for each GSH level. In conclusion, investigators proposed that "the penultimate and ultimate mutagens of 1,3-D metabolism are the corresponding epoxides and their direct hydrolysis product 3-chloro-2-hydroxypropanal, respectively." Indeed, data here show that the epoxides (or derivatives thereof) are effective mutagens, and that 3-chloro-2-hydroxypropanal (not tested in this study for mutagenicity) is a significant metabolite of the epoxides in pH 7.4 buffer. This study provides useful mechanistic information, but is not "reviewable" because only summary data were provided. There is no DPR worksheet. Aldous, 4/29/15.

50046-120 162474 Stott, W. T. and H. S. Stewart, "Determination of glutathione transferase activities in several mammalian cell lines", The Dow Chemical Co., Midland, 8/27/96. Laboratory Project Study ID # T2.06-001-014-001. Investigators evaluated activities of glutathione transferase using 4 substrates: racemic ^{14}C-UL-1,3-dichloropropene; 4-chloro-1,3-dinitrobenzene (CDNB); *p*-nitro-phenethylbromide (NPEB); and trans-4-phenyl-3-buten-2-one (TPBO). Sources of GSH transferase activities were rat liver cytosol, mouse liver cytosol, primary rat hepatocytes, CHO cell line, and two Chinese hamster lung cell lines. GSH transferase activities were compared with previously published activities for *Salmonella typhimurium*. GSH transferase activities using ^{14}C-UL-1,3-dichloropropene were about 10-fold higher for the liver 100,000 x g cytosol (rat slightly more active than mouse) compared to preparations from the three cell lines, and over 1000 times higher than was reported for the bacterial cytosol. Aldous, 9/23/99 (no worksheet).

50046-111 161854 Exact duplicate of 50046-120 162474, above.

Creedy, C. L., Brooks, T. M., Dean, B. J., Hutson, D. H., and Wright, A. S. 1984. "The protective action of glutathione on the microbial mutagenicity of the Z- and E-isomers of 1,3-dichloropropene," *Chem Biol Interact* 50:39-48. The Z(cis)- and E(trans)-isomers of 1,3-dichloropropene (DCP) were tested with TA 100 at 10 to 2000 µg/plate, either alone, with rat liver S9, with 5 mM GSH, or with S9 plus GSH. There were robust dose-related increases in revertants with both DCP isomers to the limit of cytotoxicity with DCP alone or with DCP plus S9 in the absence of GSH fortification. The presence of GSH (with or without S9) greatly reduced revertant numbers, although at dose levels of 500 to 2000 µg/plate there was perceptible response with E-DCP, and more so with Z-DPC. Investigators noted that rat liver cytosol contained somewhat more glutathione than *S. typhimurium* cytosol, and that the percentage of reduced GSH was much higher in the rodent. Also, native activities of GSH transferase activities toward Z-DCP and other GSH substrates were about 100x higher in rat liver than in bacterial cytosol. This is useful supplementary information, but not suitable for DPR review, since only summary data were provided. Aldous, 4/24/15.

Neudecker, T., and Henschler, D. 1986. "Mutagenicity of chloroolefins in the Salmonella/mammalian microsome test. III. Metabolic activation of the allylic chloropropenes allyl chloride, 1,3-dichloropropene, 2,3-dichloro-1-propene, 1,2,3-trichloropropene, 1,1,2,3-

tetrachloro-2-propene and hexachloropropene by S9 mix via two different metabolic pathways," *Mutat Res* 170:1-9. Authors investigated metabolism and mutagenicity associated with the above compounds, including the influences of S9, GSH, and particularly of key metabolic inhibitors such as SKF 525 (to inhibit microsomal oxygenases); 1,1,1-trichloropropene-2,3-oxide (TCPO: an inhibitor of epoxide hydrolase); and of cyanamide (CA: an inhibitor of aldehyde dehydrogenase). 1,3-dichloropropene 1,3-DCP was purified by vacuum distillation, and evidently contained no stabilizer. Key findings with 1,3-DCP led investigators to conclude that the dominant route of metabolism for this compound did not involve epoxide formation, but rather formation of an allyl alcohol, which could subsequently oxidized to a reactive allyl aldehyde, and eventually to the less mutagenic carboxylic acid. Mutagenicity of trans-1,3-DCP was elevated by S9, and this effect was enhanced by addition of CA (evidently by blocking oxidation of the reactive allyl aldehyde). Prolonged incubation of both cis- and trans-1,3-DCP (from 20 min to 120 min) enhanced mutagenicity. Cis- was inherently more mutagenic than trans-1,3-DCP in all cases. In most cases, mutagenicity was roughly proportional to amount of S9 present. Mutagenicity with trans-1,3-DCP was not affected by addition of SKF 525 or TCPO: this was taken to indicate that epoxide formation was not a major feature of 1,3-DCP metabolism. This study contains supplementary information, but provided only summary data, and is not suitable for a DPR worksheet. Also, unlike Schneider et al. (1998, also discussed in this section of the Toxicology Summary), the present study did not assay proposed intermediates. Aldous, 4/30/15.

Neudecker, T., Stefani, A., and Henschler, D. 1977. "In vitro mutagenicity of the soil nematicide 1,3-dichloropropene," *Experientia* 33:1084-1085. The cis- and trans-isomers of 1,3-dichloropropene were tested in the Ames mutagenicity assay system on Salmonella typhimurium tester strain TA 1535. Purity was 99.97% (cis) and 97.46% (trans). Both isomers were clearly mutagenic without S9. Survival was limiting in the range of 0.5 µl/ml top agar. S9 activation rendered test articles slightly less cytotoxic, and elicited fewer revertants (particularly where cytotoxicity was not limiting) than plates without S9. This is a very short publication, and does not require a DPR worksheet. Aldous, 4/29/15.

Mechanistic studies examined by DPR Risk Assessment Group

(Following are principally **abstracts** to published articles which were not assigned to Human Health Assessment Branch Data Review Group for examination. Individual data and quality assurance oversight do not pertain to these data, so that detailed reviews are not possible).

Watson, W. P., Brooks, T. M., Huckle, K. R., Hutson, D. H., Lang, K. L., Smith, R. J., and Wright, A. S. 1987. "Microbial mutagenicity studies with (Z)-1,3-dichloropropene," *Chem Biol Interact* 61:17-30. This study has confirmed that the direct mutagenicity previously observed when S. typhimurium TA100 was treated with (Z)-1,3-dichloropropene (DCP) was in fact due to trace impurities. These impurities result from autoxidation of (Z)-1,3-DCP and have now been identified. Both (Z)- and (E)-2-chloro-3-(chloromethyl)oxiranes (DCP oxides) were identified as significant products during this autoxidation. The mutagenic impurities formed by autoxidation were completely removed by adsorption chromatography on silicic acid. (Z)-1,3-DCP purified in this way had no direct-acting mutagenicity towards S. typhimurium TA100. However, (Z)-1,3-DCP undergoes mono-oxygenase-catalyzed conversion into bacterial mutagens in the presence of S9 fraction or washed microsomes from rat liver. The glutathione-linked conjugation systems of mammalian tissues provided efficient protection against this indirect mutagenic action.

However, the low concentration of glutathione in standard bacterial mutagenicity assays limits the glutathione S-alkyl transferase-catalyzed detoxification of (Z)-1,3-DCP and its primary bioactivation product(s). When the concentration of glutathione was adjusted to the normal physiological concentration, the mono-oxygenase-dependent mutagenic action of (Z)-1,3-DCP was virtually eliminated. These results therefore are consistent with the view that bacterial mutation assays are only qualitative indicators of potential mammalian genotoxicity.

Stolzenberg, S. J., and Hine, C. H. 1979. "Mutagenicity of halogenated and oxygenated three-carbon compounds," *Journal of toxicology and environmental health* 5:1149-1158.
Four structurally related three-carbon compounds, known for their antifertility activity in the male, and the brominated derivatives of two of these compounds were tested for mutagenic activity by the Salmonella typhimurium test of Ames et al. In the presence of strain TA-100, a base-pair substitution detector strain, 1,2-dibromo-3-chloropropane (DBCP), was the most active compound tested but required enzymatic conversion by S9 microsomal preparation to an active mutagen. Three of these compounds containing an epoxide group - epichlorohydrin, epibromohydrin, and glycidol - were highly active direct mutagens, not requiring S9 for activation. α-Chlorohydrin was the least active compound tested; α-bromohydrin was 40 times more active than its chlorinated analog. Epibromohydrin was only slightly more active than epichlorohydrin, but both were highly active. With both of the halogenated epoxides, S9 preparation caused a substantial decrease in mutagenic activity at every concentration tested. All six compounds showed dose-related responsiveness for the base-pair substitution detector strains used. However, they were relatively inactive against the frameshift detector strain of S. typhimurium, TA-98. Glycerol, propylene glycol, and n-propanol, which are also three-carbon compounds containing one or more hydroxy groups, were inactive when tested at high concentrations with strain TA-100.

Talcott, R. E., and King, J. 1984. "Mutagenic impurities in 1,3-dichloropropene preparations," *Journal of the National Cancer Institute* 72:1113-1116.
A widely used pesticide, 1,3-dichloropropene [(DCP) CAS: 542-75-6], has been reported to be mutagenic to Salmonella typhimurium TA100, but large variations in specific mutagenic activity have been observed among different preparations. The purposes of this investigation were to determine the probable cause of the interpreparational variation and to provide new information on the nature of the mutagenic activity. Four preparations were assayed for mutagenic activity before and after silicic acid chromatography. None of the preparations retained mutagenic activity after chromatography, but each contained direct-acting mutagenic polar impurities. The specific mutagenic activities of the unpurified DCP samples appeared to be determined by the mutagenic activities of their polar impurities. A mixture of mutagenic polar impurities could be regenerated by refluxing a purified DCP preparation for 6 hours. The fraction of polar impurities from one of the preparations was analyzed by gas chromatography-mass spectroscopy. Although its composition was too complex to characterize completely, two known mutagens, epichlorohydrin (CAS: 106-89-8; 1-chloro-2,3-epoxypropane) and 1,3-dichloro-2-propanol (CAS: 96-23-1), were tentatively identified. In view of these results, future studies are required to establish whether DCP itself is a chemical carcinogen or whether its previously observed carcinogenicity resulted from the presence of mutagenic impurities.

Martelli, A., Allavena, A., Ghia, M., Robbiano, L., and Brambilla, G. 1993. "Cytotoxic and genotoxic activity of 1,3-dichloropropene in cultured mammalian cells," *Toxicology and Applied Pharmacology* 120:114-119.
1,3-Dichloropropene (DCP), a widely used soil fumigant previously found to be carcinogenic in both mice and rats, was evaluated for its cytotoxic and genotoxic effects in cultured rodent and human cells. A reduction of cell viability that was dependent on the dose and the length of treatment was observed with the trypan blue and the neutral red assay in both V79 cells and rat hepatocytes exposed to DCP concentrations ranging from 0.18 to 5.6 mM. In the absence of a metabolic activation system, a dose-dependent frequency of DNA single-strand breaks, that were only partially repaired within 24 hr, was revealed by the alkaline elution technique in V79 cells exposed to sub-toxic DCP concentrations. The genotoxicity of DCP was confirmed by the results obtained in metabolically competent primary cultures of both rat and human hepatocytes which displayed similar dose-related amounts of DNA fragmentation and DNA repair synthesis, and showed, in comparison to metabolically deficient V79 cells, a somewhat greater sensitivity to the cytotoxic and DNA damaging effects of DCP. The increase in the frequency of DNA breaks observed in rat hepatocytes after GSH depletion confirms the role of this tripeptide in DCP detoxification; its reduction in hepatocytes simultaneously exposed to metyrapone is consistent with a cytochrome P450-dependent biotransformation of DCP to more toxic metabolites.

Loveday, K. S., Lugo, M. H., Resnick, M. A., Anderson, B. E., and Zeiger, E. 1989. "Chromosome aberration and sister chromatid exchange tests in Chinese hamster ovary cells in vitro: II. Results with 20 chemicals," *Environ Mol Mutagen* 13:60-94.
Twenty chemicals were tested for their ability to induce sister chromatid exchanges (SCEs) and chromosomal aberrations (ABs) in cultured Chinese hamster ovary cells (CHO). These chemicals were tested with and without an added metabolic activation system (rat liver S9 fraction). Four chemicals were negative in both assays, 1 induced ABs only, and 15 were positive for SCEs; 6 of these 15 also induced ABs. The effect of cell harvest time on the ability to detect the induction of chromosomal aberrations was examined for six chemicals. Five of these had caused at least one of the following: cell cycle delay, aberrations observed in first division metaphase cells in the SCE assay, or a weak response in the standard AB assay (10-12-hr growth period). Three chemicals, chlorinated trisodium phosphate, 1,2-dibromo-3-chloropropane, and tetrakis(hydroxymethyl)phosphonium chloride, were positive using both the standard and extended harvest times. N-Nitrosodimethylamine and diphenhydramine HCl were only positive using an extended harvest time, and malonaldehyde was negative using both standard and extended harvest times.

Ghia, M., Robbiano, L., Allavena, A., Martelli, A., and Brambilla, G. 1993. "Genotoxic activity of 1,3-dichloropropene in a battery of in vivo short-term tests," *Toxicology and applied pharmacology* 120:120-125.
The genotoxic activity of 1,3-dichloropropene, which has been classified as possibly carcinogenic to humans, was investigated in rats given high single doses of this chloroolefin. A dose-related amount of DNA fragmentation was observed at doses ranging from 62.5 to 250 mg/kg in liver and gastric mucosa, both of which are targets of DCP carcinogenic activity, as well as in the kidney. The frequency of DNA breaks, that were to a large extent repaired within 24 hr, was higher after po than after ip administration in the liver, while the converse occurred in the kidney. Any evidence of DNA fragmentation was, in contrast, absent in lung, bone marrow,

and brain which are not sites of DCP-induced tumor development. A role of cytochrome P450 in the activation of DCP is suggested by the lower degree of liver DNA fragmentation observed in rats pretreated with methoxsalen. DCP produced a dose-dependent reduction of the liver GSH level, an effect that presumably hinders its detoxification and thus favors its DNA-damaging activity. In contrast with the satisfactory prediction of DCP carcinogenic activity provided by the results of the in vivo DNA damage/alkaline elution assay, neither the in vivo rat hepatocyte DNA repair assay nor the micronucleus assay, carried out on bone marrow, spleen, and liver cells of partially hepatectomized rats, supplied any evidence of DCP genotoxicity. NOTE: (Aldous, 6/24/15), in this study, DCP was dissolved in DMSO immediately prior to dosing. Schneider, Quistad, and Casida (1998) had reported that DMSO elicited a spontaneous degradation of DCP toward 2-chloroacrolein, which is not a common metabolite under physiological conditions.

Kevekordes, S., Gebel, T., Pav, K., Edenharder, R., and Dunkelberg, H. 1996. "Genotoxicity of selected pesticides in the mouse bone-marrow micronucleus test and in the sister-chromatid exchange test with human lymphocytes in vitro," *Toxicology letters* 89:35-42.
Selected pesticides (aldicarb, 1,3-dichloropropene, methidathion, parathion, triadimefon, vinclozolin) were tested for their clastogenic and aneugenic activities in the mouse bone-marrow micronucleus (MN) test in vivo and for their sister-chromatid exchange-inducing activities in human lymphocytes in vitro in the presence and absence of an exogenous metabolizing system from rat liver S9. 1,3-Dichloropropene significantly increased the frequencies of micronucleated polychromatic erythrocytes (PCE) in bone-marrow cells of female mice from 3.3 MN/1000 PCE to 15.3 MN/1000 PCE (187 mg per kg body weight). 1,3-Dichloropropene (100 μM) induced 16.0 SCE/metaphase after 24 h of incubation as compared with the basal rate of 11.2 SCE/metaphase (- S9) and of 15.4 SCE/metaphase as compared with 10.5 SCE/metaphase of the control (+ S9). These values were statistically significantly different from each other. The other pesticides tested did neither increase the rate of micronuclei significantly in polychromatic erythrocytes in male nor in female animals. Aldicarb and methidathion induced a significant increase in SCEs in human lymphocytes in vitro only without the metabolic activating system: aldicarb, 5 μM, 24 h incubation: 15.5 SCE/metaphase; control: 12.6 SCE/metaphase; methidathion, 100 μM, 24 h incubation: 15.8 SCE/metaphase, control: 11.1 SCE/metaphase. Parathion, triadimefon and vinclozolin did not have any SCE-inducing effects. NOTE: (by Aldous, 6/24/15), DCP was 95% purity, and was administered in corn oil for in vivo studies. Investigators have no explanation for why females elicited a strong response in vivo and in vitro for micronucleated PCE's, whereas males had no response at all.

Schiffmann D, Eder E, Neudecker T, & Henschler D (1983) "Induction of unscheduled DNA synthesis in Hela cells by allylic compounds," Cancer Lett, 20(3): 263-269. Thirteen allylic compounds, mostly with close structural relationship, were tested for their ability to induce unscheduled DNA synthesis (UDS) in HeLa cells and mutations in the Ames test; 11 induced UDS in dose dependence. Allyl isothiocyanate was negative in UDS (borderline in the Ames test) and acrolein (positive in the Ames test) proved toxic to HeLa cells, therefore UDS measurement was excluded. In general, positive qualitative and quantitative correlation between UDS, Ames test and alkylating properties 9as measured in the 4-nitrobenzyl-pyridine test, NBP) were found. Among structural analogs and typical allylic compounds with various leaving groups, the amount of induced DNA repair at equimolar concentrations decreased in the same order as the mutagenic and alkylating activities in the other 2 test systems: 1,3-dichloropropene

(cis) > 1,3-dichloropropene (trans) > 2,3,-dichloro-1-propene; 1-chloro-2-butene > 3-chloro-1-butene > 3-chloro-2-methyl-1-propene > allyl chloride; allyl-methanesulfonate > -iodide > -bromide > -chloride.

von der Hude, W., Scheutwinkel, M., Gramlich, U., Fissler, B., and Basler, A. 1987. "Genotoxicity of three-carbon compounds evaluated in the SCE test in vitro," *Environmental mutagenesis* 9:401-410. Sister chromatid exchange increased with dose of 1,3-dichloropropene, to the limits of cytotoxicity. This study used DMSO as a vehicle. As noted elsewhere in this Summary of Toxicology Data, Schneider, Quistad, and Casida (1998) had reported that DMSO elicited a spontaneous degradation of DCP toward 2-chloroacrolein, which is not a common metabolite under physiological conditions. As a result, this study may not reflect a plausible exposure scenario. No individual data were provided. As there are reviewable reports consistent with the key conclusions already available, this report does not warrant further examination. Aldous, 6/24/15.

Vithayathil, AJ; McClure, C; Myers, JW. (1983) "Salmonella/microsome multiple indicator mutagenicity test," Mutat Res 121(1):33-37. This article primarily seeks to explore the benefits of using a single Salmonella strain (TA98) to address frameshift mutations via histidine backwards mutation as well as rifampicin resistance via a forward base-pair substitution mutation. Several test compounds, including 1,3-dichloropropene, were positive in both respects, suggesting some measure of base-pair and frameshift mutation potential. No individual data were provided. As there are reviewable reports consistent with the key conclusions already available, this report does not warrant further examination. Aldous, 6/24/15.

Stolzenberg, S. J., and Hine, C. H. 1980. "Mutagenicity of 2- and 3-carbon halogenated compounds in the Salmonella/mammalian-microsome test," *Environmental mutagenesis* 2:59-66. This article compared mutagenic potencies of various bromo- and chloro- substituted molecules, including 1,3-dichloropropene, with respect to strain TA-100, with and without S-9. Data for 1,3-dichloropropene demonstrate a mutagenic response, with far more revertants in plates without S-9 at any given dose level. No individual data were provided. As there are reviewable reports consistent with the above observation already available, this report does not warrant further examination. Aldous, 6/24/15.

Haworth, S; Lawlor, TK; Mortelmans, K; et al. (1983) "Salmonella mutagenicity testing for 250 chemicals," Environ Mutagen Suppl 1:3-142. This article codes results from many laboratories for 250 chemicals as "+" or "-." The entry in the table for 1,3-dichloropropene has a "+" for mutagenicity (not otherwise specified) from a study undertaken by SRI. As there are reviewable reports (positive and negative for mutagenicity) already available, this reference does not warrant further examination. Aldous, 6/24/15.

Registrant interpretive commentary and EPA documents

50046-094 138953 DowElanco reported as FIFRA 6(a)(2) "possible adverse effect" data on 6/20/95 that they had learned that a European 1,3-dichloropropene product (using soybean epoxide as stabilizer, but not a product sold in the United States) elicited stomach lesions considered to be pre-neoplastic in subchronic studies. The memo noted that Sprague-Dawley rats administered 25 mg/kg/day or more daily for 28 days had squamous cell hyperplasia and hyperkeratosis in the forestomach (NOEL = 5 mg/kg/day). Also, CD-1 mice had mild cases of

hyperplasia and hyperkeratosis in the forestomach at 200 mg/kg/day after 28 days of treatment (lower doses were included in the study, but not evaluated as of the time of the memo). One of 3 attachments was not otherwise included in the present Summary of Toxicology Data: a Dec. 8, 1989 peer review of Telone II cited several tumor types observed following Telone treatment, including forestomach tumors. One contributing document to that peer review was the May 1985 NTP study employing F344 rats and B6C3F1 mice, both of which acquired forestomach tumors, as noted elsewhere in this Summary. No DPR review, since there were no fundamentally new reviewable data. Aldous, 9/9/99.

50046-007, 932850; Communication to EPA from Dow dated 2/9/82; contains risk assessment based on data from NTP rat and mouse studies (# 036552 & 53) as well as published dermal studies (# 036554), and refers to oncogenic effects noted in the former. (Martz, 8/18/86).

50046-016, 932849, 932853, and 022757; Contain preliminary summary of NTP studies (# 036552 & 53), summary of mutagenicity studies showing positive effects (# 036556-58), and summary of the one generation reproduction study with technical D-D (# 36555), respectively. (Martz, 8/18/86).

50046-016 149370 Brief summaries of toxicology data as of 1982. No reviewable data. Aldous, 3/10/97

50046-139 169270 Rao, K. S., "Telone II: 1,3-dichloropropene (1,3-D): mammalian risk assessment", Final Draft Document submitted to Toxicology Excellence for Risk Assessment (TERA) panel members, Dow AgroSciences, 10/20/98. **Summary makes the following assertions and recommendations**: current production Telone II lacks mutagenic potential under normal physiological conditions. Older formulations had epichlorohydrin as a stabilizing agent, which evidently contributed to mutagenic and oncogenic properties of that material. Current production Telone II lacks epichlorohydrin, and also the associated risks. Dose levels of Telone II high enough to deplete tissue levels of glutathione may elicit oncogenicity (as in rat liver). Often the primary chronic findings are related to local irritant actions of Telone II to mucosal tissues as determined by route (as to the forestomach lining or respiratory tract). Results from exposures to very high dose levels are not representative of plausible human exposures. This assessment concluded that benchmark methodology at the 0.1 level could be used to derive RfC values of 41 $\mu g/m^3$ for chronic toxicity for mouse nasal epithelial effects, or 80 to 800 $\mu g/m^3$ for bronchioalveolar adenomas; and RfD values of 0.022 mg/kg/day for male rat forestomach hyperplasia and 0.025 to 0.008 mg/kg/day for liver tumors. If cancer potency data are required to be used based on oncogenicity results, the potency calculations would be 1.3×10^{-5} $(\mu g/m^3)^{-1}$ and 4.0×10^{-2} $(mg/kg/day)^{-1}$, respectively.

The above record continues with attachments, as numbered below:

(1) Several communications dating up to 1993 relating to status of Telone II with the European Community partners.

(2) IRIS record for 1,3-dichloropropene dating to 1993.

(3) U.S. EPA Reregistration Eligibility Document (2 Sept. 1997). This document noted that the negative results in the recent inhalation-route dominant lethal assay (Record No. 162466, above) "... lessen the concern for germ cell [mutagenic] effects; therefore, no further

mutagenicity testing is required" (p. 10). The HED derived a Q_1* value of 5.33 x 10^{-2} in 1994 based on male mouse bronchioalveolar adenomas (p. 13). When the HED RfD Peer Review Committee met in January, 1997 to re-evaluate status of Telone II, they determined that there was no need to change the "carcinogenicity classification" nor the Q_1* (p. 14), largely due to positive mutagenicity studies which support the "weight-of-evidence" for Telone II as an oncogen.

(4, 5, 6, and 7) These four attachments provide the mathematical analyses for "point of departure" evaluations. For non-cancer effects, Attachment 4 relates to female mouse nasal epithelial hyperplasia and hypertrophy following inhalation exposure (pp. 55 ff. of report), and Attachment 5 relates to male rat forestomach non-glandular mucosal basal cell hyperplasia (pp. 59 ff. of report). For cancer effects, Attachment 6 relates to inhalation exposure in the mouse oncogenicity study (Record No. 060675), in which male mice had elevated bronchioalveolar adenomas at the highest dose level only. This document proposes a benchmark concentration model (p. 62 of report), however linearized multistage extrapolation analyses are also provided (p. 66 of report). The final cancer evaluation (Attachment 7) relates to male rat liver tumors following oral exposure. Both benchmark and linearized multistage extrapolation analyses were discussed in the report (pp. 68 to 72). Attachments 6 and 7 provided strictly benchmark approach analyses.

Attachments 8 and 9 address dosimetry of inhaled toxicants with respect to respiratory tract anatomy and physiology in order to extrapolate animal data to humans.
The above data do not provide data appropriate for review under SB-950. No worksheet. Aldous, 7/22/99.

50046-116 162467 DowElanco response to draft HED and EFED RED chapters (relates to 1997 U.S. EPA document on 1,3-dichloropropene). Primary interest for this Summary is the tab: "Response to Tox. Portions - Draft HED RED Chapter". Authors (Stott, W. T. and B. B. Gollapudi) determined that the EPA document made excessive use of outdated studies and applied highly conservative risk extrapolation models. Further, recent mammalian metabolism and mutagenicity studies were often ignored in favor of older *in vitro* studies, hence results often do not have relevance to physiological responses of plausible exposure scenarios. Toxicity studies selected for analyses by U.S. EPA were often bolus-dose treatments, which are known to be able to saturate normal physiological defense mechanisms. Many older studies used obsolete formulations of 1,3-dichloropropene containing up to 2% epichlorohydrin, which is a known mutagenic stabilizing agent not found in current production. Many of the lesions, including tumors, elicited by 1,3-dichloropropene are port-of-entry effects which would not be expected to occur under most plausible exposure scenarios. No worksheet (no "reviewable" data). Aldous, 7/26/99.

50046-117 162468 This is a continuation of 162467, by the same authors. Primary contribution is evidence that 1,3-dichloropropene does not bind to calf thymus DNA *in vitro* nor in liver nor lungs of F344 rat nor in B6C3F1 mice *in vivo*. Liver GSH depletion was shown at gavage doses of 25 to 100 mg/kg/day for 3 days (NOEL = 12.5 mg/kg/day). Lung GSH was dose-related in the range of 10 to 150 ppm (NOEL not sought nor obtained in this study). See Document No. 50046-119 for details. No worksheet for this brief summary record. Aldous, 7/26/99.

50046-118 162469 Calhoun, L. L. "Additional comments in response to the risk assessment portions of the draft HED RED chapter for 1,3-D: November 21, 1997." This brief record states that threshold-based calculations should be used for chronic and oncogenicity findings. Tables therefore present benchmark dose (BMD) analyses for male rat forestomach basal cell hyperplasia and for male rat hepatocellular adenoma. No worksheet (no "reviewable" data). Aldous, 7/26/99.