CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

Aminocyclopyrachlor

Chemical Code # 5998, **Tolerance #** 53081

Original: April 23, 2009
Revised: July 15, 2010
Revised: August 8, 2011

I. DATA GAP STATUS

Chronic toxicity, rat:	No data gap, no adverse effect indicated.
Chronic toxicity, dog:	No data gap, no adverse effect indicated.
Oncogenicity, rat:	No data gap, no adverse effect indicated.
Oncogenicity, mouse:	No data gap, no adverse effect indicated.
Reproduction, rat:	No data gap, no adverse effect indicated.
Teratology, rat:	No data gap, no adverse effect indicated.
Teratology, rabbit:	No data gap, no adverse effect indicated.
Gene mutation:	No data gap, no adverse effect indicated.
Chromosome effects:	No data gap, no adverse effect indicated.
DNA damage:	No data gap, no adverse effect indicated.
Neurotoxicity:	Not required at this time

Toxicology one-liners are attached.

All record numbers through 259382 were examined.
** indicates an acceptable study.
Bold face indicates a possible adverse effect.

File name: T110808
Prepared by T. Moore 8/8/11

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

These pages contain summaries only. Individual worksheets may contain additional effects.

COMBINED, RAT

** 53146-0002; 259016; "DPX-MAT28 Technical: Combined Chronic Toxicity/Oncogenicity 2-Year Feeding Study in Rats"; (K.S. Moon; Korea Institute of Toxicology, 100 Jang-dong, Yu Seong-gu, DaeJeon 305-600, Republic of Korea; Study No. IG07281; 11/24/10); Seventy Crl:CD (SD) rats/sex/group received 0, 600, 2000, 6000 or 18000 ppm of DPX-MAT28 Technical (aminocyclopyrachlor potassium salt) (lot no. DPX-MAT28-023; purity: 90.5% (8/30/07), revised to 88.3% (12/3/08)) in the diet for 2 years ((M) 0, 27.4, 97.1, 278.6, 892.2 mg/kg/day, (F) 0, 29.3, 99.8, 308.7, 957.3 mg/kg/day). An additional cohort of 10 animals/sex/group were treated for one year with the test material and included in the chronic toxicity evaluation. There was no treatment-related effect on the survival of the study animals. The mean body weight gain of both sexes in the 18000 ppm group was less than the control values. Although the mean food consumption for both sexes in the high dose group was less than that of the control group at various times during the study, there was no substantive difference in the overall consumption on a per-animal basis. There were no apparent treatment related effects on the eyes of the study animals of either sex. Hematology parameters were not affected by the treatment. In the clinical chemistry evaluation, the serum globulin levels for both sexes in the 18000 ppm group were less than those of the control group after 6 months of treatment ($p<0.05$ or 0.01). In the urinalysis, the mean pH value of the 18000 ppm females was less than that of the control group after 6 months of treatment ($p<0.01$). The pH values of the urine for both sexes in the 18000 ppm were also less than the mean values for the control group (NS). In the histopathological evaluation, an increased incidence of astrocytomas were noted in the brain of the 18000 ppm males (0: 0/70 vs. 6000: 1/70, 18000: 3/69 ($p<0.05$)). In addition, when combined glial neoplasms were considered, the incidence was as follows: (0: 0/70 vs. 6000: 2/70, 18000: 4/69 ($p<0.05$)). DuPont convened a Pathology Working Group (PWG) to evaluate the toxicological significance of these observations. The PWG concluded that the lesions were not treatment-related for the following reasons: 1) The incidences of both the astrocytomas and the total glial neoplasms were within the historical control range (observed incidence: astrocytoma, 4.4% (18000 ppm group), combined glial, 5.8% (18000 ppm group). 2) Gliosis was not evident as a non-neoplastic precursor. 3) The incidence of astrocytoma or combined glial neoplasms was not noted as a treatment-related effect in the females even though the pharmacokinetics of the test material was similar for both sexes. 4) There was no apparent decrease in latency of onset for the brain tumors with an increase in treatment level. 5) There are no other oncogenic effects evident. 6) No positive assays were reported for the genotoxicity testing. **No adverse effect indicated. Rat Chronic Toxicity Dietary NOEL:** (M/F) 6000 ppm (based upon treatment-related effects upon the body weight gain of both sexes in the 18000 ppm group) ((M) 278.6 mg/kg/day, (F) 308.7 mg/kg/day); **no oncogenicity was evident. Study acceptable.** (Moore, 6/6/11)

CHRONIC TOXICITY, RAT

See Combined, Rat above.

CHRONIC TOXICITY, DOG

** 53146-0003; 259017; "DPX-MAT28 Technical: Chronic Oral Toxicity One-Year Feeding Study in Beagle Dogs"; (S.C. Han; Korea Institute of Toxicology, 100 Jang-dong, Yu Seong-gu, DaeJeon 305-600, Republic of Korea; Study No. IG07380; 10/22/10); Four beagle dogs/sex/group received 0, 1250, 5000, 15000 or 30000 ppm of DPX-MAT28 Technical (aminocyclopyrachlor potassium salt) (lot no. DPX-MAT28-023; purity: 90.5% (8/30/07), revised to 88.3% (12/3/08)) for 12 months ((M) 0, 38.1, 179.0, 467.7, 1082.7 mg/kg/day, (F) 0, 0, 47.1, 175.6, 545.1, 1078.4 mg/kg/day). No unscheduled deaths occurred during the study. There was no treatment-related effect upon the mean body weight gain or food consumption. The hematology evaluation did not demonstrate any treatment-related effects. No treatment-related

effects were evident in the clinical chemistry evaluation. The ophthalmology examination did not reveal any treatment-related lesions. In the urinalysis, the pH values for the females in the 30000 ppm group were generally lower than the control values over the course of the study. The mean absolute and relative liver weights of the 30000 ppm females were greater than the control values (NS). In the histopathology examination, there was an increased incidence of vacuolation in the livers of the 30000 ppm females (0: 0/4 vs. 1250: 1/4, 5000: 0/4, 15000: 1/4, 30000: 2/4). **No adverse effect indicated. Dog Chronic Dietary Toxicity NOEL:** (M) 30000 ppm (1083 mg/kg/day) (based upon the lack of treatment related effects upon the males in the 30000 ppm group), (F) 15000 ppm (545 mg/kg/day) (based upon the vacuolation noted in the liver of the females in the 30000 ppm group); **Study acceptable.** (Moore, 6/7/11)

ONCOGENICITY, RAT

See Combined, Rat above.

ONCOGENICITY, MOUSE

** 53146-0001; 259015; “DPX-MAT28 Technical: Oncogenicity 18-Month Feeding Study in Mice”; (Huh, J.I; Korea Institute of Toxicology, 100 Jang-dong, Yu Seong-gu, DaeJeon 305-600, Republic of Korea; Study No. IG07280; 10/15/10); Sixty Crlj: CD1 mice/sex/group received 0, 300, 1000, 3000 or 7000 ppm of DPX-MAT28 Technical (aminocyclopyrachlor potassium salt) (lot no. DPX-MAT28-023; purity: 90.5% (8/30/07), revised to 88.3% (12/3/08)) in the diet for 18 months ((M) 0, 38.7, 132.7, 393.1, 876.2 mg/kg/day, (F) 0, 49.9, 170.6, 526.8, 1190 mg/kg/day). There was no treatment-related effect on survival. Mean body weight and food consumption were not affected by the treatment. The ophthalmological evaluation did not reveal any treatment-related effects. The differential distribution of the white blood cell types was not affected by the treatment. The mean absolute and relative organ weights were not affected by the treatment. The incidence of oncogenic lesions was not treatment-related. **No adverse effect was evident. Mouse Chronic Dietary NOEL:** (M/F) 7000 ppm (M: 876 mg/kg/day, F: 1190 mg/kg/day) (based on the lack of treatment-related effects for both sexes in the 7000 ppm group); **no oncogenicity was evident. Study acceptable.** (Moore, 6/1/11)

REPRODUCTION, RAT

**53081-0081 243089, “DPX-MAT28 Technical: Multi-generation Reproduction Study in Rats”, (J. M. Lewis, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID. DuPont-22032, 15 October 2008). 28 Crl:CD(SD) rats per sex per group received DPX-MAT28 Technical (90.9%) in the diet at 0 (basal diet), 500, 1500, 5000, and 17000 ppm through 2 generations with 1 litter per generation. Group mean daily intake of DPX-MAT28 Technical for F0 and F1 males was 0, 30.11, 91.93, 299.09, and 1048.26 mg/kg/day and 0, 42.32, 126.33, 425.76, and 1521.77 mg/kg/day at 0, 500, 1500, 5000, and 17,000 ppm, respectively. Group mean daily intake for F0 and F1 females was 0, 35.97, 110.02, 367.0, and 1243.26 mg/kg/day and 0 46.42, 140.93, 465.20, and 1665.55 mg/kg/day at 0, 500, 1500, 5000, and 17,000 ppm, respectively, during the premating period. Significant reductions in group mean bodyweight and bodyweight gains were noted for F0 males and females (during premating, gestation, and/or lactation) at 17,000 ppm. Bodyweights and bodyweight gains were 6% to 9% and 13% to 17% lower respectively for F0 males vs control values. F0 female bodyweights were 6% to 7% lower during premating, 8% lower during gestation, and 5% to 9% lower during lactation, while bodyweight gain decreases for the corresponding periods were 16% (premating), 12% (gestation), and 0 (gains were noted for the lactation period, 13.5 g vs 0.5 g for controls). At 5000 ppm, bodyweights and bodyweight gains were significantly reduced (6% to 7% and 14% lower than controls, respectively) for F0 males and comparable to controls for F0 females. At 17,000 ppm, group mean bodyweight and bodyweight gain were significantly reduced for F1 males (12% to 15% decreases for both parameters) and F1 females (reductions of 14% for bodyweight and 5% for bodyweight gain during premating) compared to controls. At 5000 ppm, significant reductions in bodyweight and bodyweight gain (7% to 9%) were noted for F1 males (values for F1 females were generally comparable to controls. No treatment-related clinical observations were recorded for F0 and F1 animals during the study. F0 and F1 parameters for sperm, estrous cycle, mating, fertility, pre-

coital interval length, gestation length, implantation site counts, and post-implantation losses were not effected by treatment. There were no treatment-related changes to F1 female ovarian follicle counts. Microscopy revealed an increase in thyroid follicular cell hypertrophy (grade 1 (minimal) characterized by an increase in the size of the follicular lining cell and a change in shape from flat or cuboidal to columnar) for F1 adult females at 17,000 ppm compared to controls (2/28 vs 8/28). Group mean F1a pup weights were significantly reduced at 5,000 ppm (7% lower than controls on lactation day 21) and 17,000 ppm vs controls (group mean pup weights were 7%, 7%, 8%, and 15% lower than controls on lactation days 4, 7, 14, and 21, respectively). F2a pup weights were significantly decreased vs controls (8%) at 17,000 ppm on lactation day 21. F1a and F2a group mean relative spleen weights were reduced (statistically significant in F1a pups) compared to controls at 17,000 ppm. Preputial separation and vaginal patency in F1a offspring were not effected by treatment. Parental NOEL based on reduced bodyweight and bodyweight gain: Males = 1500 ppm (F0 males, 91.93 mg/kg/day and F1 males, 126.33 mg/kg/day) and Females = 5000 ppm (F0 females, 367.0 mg/kg/day and F1 females, 465.2 mg/kg/day). Pup NOEL = 5000 ppm (reduced pup weights on day 21). Reproductive NOEL = No effect at the HDT. No adverse reproductive effects. Acceptable. (Green, 3/11/09).

TERATOLOGY, RAT

**53081-0080 243088, "DPX-MAT28 Technical: Developmental Toxicity Study in Rats", (J.M. Lewis, E.I. DuPont de Nemours and Company, DuPont Haskell℠ Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID. DuPont-22378, 21 February 2008). 25 time-mated female Crl:CD(SD) rats per groups received DPX-MAT28 Technical (92.2% aminocyclopyrachlor) by oral gavage at 0 (0.5% methylcellulose), 30, 100, 300, and 1000 mg/kg/day on gestation days 6 through 20. No maternal deaths occurred and no treatment-related effects were noted for clinical observations, bodyweight, food consumption, maternal necropsies and reproductive outcomes, organ weights, or implantation/fetal/litter parameters, and fetal alterations. Maternal and Developmental NOEL = 1000 mg/kg/day. No teratogenicity. Acceptable. (Green, 3/2/09).

TERATOLOGY, RABBIT

**53081-0079 243087, "A Prenatal Developmental Toxicity Study of DPX-MAT28 in Rabbits, Revision 2, Final Report", (T.L. Fleeman, WIL Research Laboratories, LLC, Ashland, OH., Sponsor study No. DuPont-22377, 14 July 2008). 22 time-mated female New Zealand White rabbits per group received DPX-MAT28 (92.2% aminocyclopyrachlor) by oral gavage at 0 (0.5% methylcellulose), 100, 300, 500, and 1000 mg/kg/day on gestation days 7 through 28. At 1000 mg/kg/day, 2 females were found dead (1 on gestation day 13 (bodyweight loss, reduced food consumption, and decreased defecation were noted in the preceding days) and the other on day 27 (intubation error)) and 2 females aborted (1 on day 20, the other on day 26 (both females exhibited decreased defecation, reduced bodyweight gain, and reduced food consumption for several days prior to abortion)). One 300 mg/kg/day female aborted on gestation day 28 (clinical signs on preceding days included decreased defecation, soft stool, and reduced food consumption) and one 100 mg/kg/day female was found dead on day 22 with no preceding clinical signs. Group mean maternal bodyweight gain was reduced at 1000 mg/kg/day during the treatment period (significant for days 14-17) relative to controls. At 500 mg/kg/day, mean maternal bodyweight loss was noted for gestation days 24-29 (ns and bodyweights were comparable to controls on the other treatment days). Group mean maternal food consumption was reduced at 500 mg/kg/day (gestation days 14-20) and 1000 mg/kg/day (gestation days 14-24). Maternal necropsy results and fetal external, visceral, and skeletal evaluation results were unremarkable. Maternal NOEL = 300 mg/kg/day (reduced food consumption and bodyweight gain). Developmental NOEL = 1000 mg/kg/day. No teratogenicity. Acceptable. (Green, 2/26/09).

GENE MUTATION

Gene Mutation

**53081-0083 243091, "DPX-MAT28 Technical: Bacterial Reverse Mutation Assay", (V.O.

Wagner and M.R. VanDyke, BioReliance, Rockville, MD, BioReliance study No. AC02HJ.503.BTL, 5 October 2007). Triplicate cultures of *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 and *Escherichia coli* strain WP2 uvrA were exposed (direct plate incorporation) to DPX-MAT28 Technical (92.2% aminocyclopyrachlor), in the presence and absence of S9 mix, at 0 (DMSO), 50, 150, 500, 1500, and 5000 ug/plate for 48 to 72 hours (37 $\pm$ 2° C). Cytotoxicity was not indicated. There were no increases in the number of revertants per plate. Positive controls were functional. Acceptable. (Green, 1/27/09).

53081-0084 243093, "DPX-MAT28 Technical: *In Vitro* Mammalian Cell Gene Mutation Test (CHO/HGPRT)", (J.J. Clarke, BioReliance, Rockville, MD, BioReliance study No. AC02HJ.782.BTL, 9 October 2007). Exponentially growing Chinese hamster ovary cells (CHO-K_1) were seeded in treatment medium at 5 x 10^5 cells/25 cm^2 flask and incubated (37 $\pm$ 1° C with 5 $\pm$ 1% CO_2 in humidified air) for 18 to 24 hours followed by exposure in duplicate to DPX-MAT28 Technical (92.2% aminocyclopyrachlor), in the presence and absence of S9 mix, at 0 (DMSO), 750, 1000, 1500, 1750, and 2150 ug/ml for 5 hours at (37 $\pm$ 1° C). No increase in forward gene mutations was indicated. Positive controls were functional. Unacceptable and not upgradeable (lack of a confirmatory trial). (Green, 4/22/09).

**53081-0097 243106, "IN-KJM44: Bacterial Reverse Mutation Test", (L. S. Ford, E.I. DuPont de Nemours and Company, Haskell Laboratory for Health and Environmental Sciences, Newark, DE., Laboratory Project ID: Dupont-17971, 18 November 2005). Triplicate cultures of *Salmonella typhimurium* strains TA98, TA100, TA1535, and TA1537 and *Escherichia coli* strain WP2 uvrA were exposed (direct plate incorporation) to DPX-IN-KJM44 Technical (95.3% aminocyclopyrachlor), in the presence and absence of S9 mix, at 0 (DMSO), 100, 333, 1000, 3333, and 5000 ug/plate for 48 to 72 hours (37 $\pm$ 2° C). There was no increase in the number of revertants per plate. Precipitation was not observed and cytotoxicity was not indicated at any concentration. Positive controls were functional. Acceptable. (Green, 2/18/09)

Chromosome Effects

**53081-0085 243094, "DPX-MAT28 Technical: *In Vitro* Mammalian Chromosome Aberration Test in Human Peripheral Blood Lymphocytes", (R. Gudi and M. Rao, BioReliance, Rockville, MD, BioReliance study No. AC02HJ.341.BTL, 5 October 2007). Duplicate cultures of human peripheral blood lymphocytes were exposed to DPX-MAT28 Technical (92.2% aminocyclopyrachlor) at 0 (DMSO), 267, 534, 1068, and 2136 µg/ml for 4 hours in the presence and absence of rat liver S9 mix, and for 20 hours in the absence of activation. Cells were harvested 20 hours after the start of treatment. 100 metaphases per duplicate culture were evaluated. No increase in structural chromosomal aberrations was indicated. At 2136 µg/ml, mitotic inhibition was 8%, 54%, and 0% compared to the solvent control for non-activated cultures treated for 4 and 20 hours and for activated cultures (4 hour treatment), respectively. Positive controls were functional. Acceptable. (Green, 2/3/09).

DNA DAMAGE

**53081-0086 243095, "DPX-MAT28 Technical: Mouse Bone Marrow Erythrocyte Micronucleus Test", (L. Krsmanovic and T. Huston, BioReliance, Rockville, MD, BioReliance study No. AC02HJ.123.BTL, 5 October 2007). 10 Crl:CD1(ICR) mice per sex per group received a single oral gavage dose of DPX-MAT28 Technical (92.2% aminocyclopyrachlor) at 0 (DMSO), 500, 1000, and 2000 mg/kg with bone marrow sampling (5 per sex) 24 and 48 hours later. Mice in all treatment groups appeared normal during the observation period. 2000 polychromatic erythrocytes per animal were evaluated and scored by light microscopy. There was no increase in micronucleated polychromatic erythrocytes. Positive controls were functional. Acceptable. (Green, 2/17/09).

NEUROTOXICITY

**53081-0070, 071 243078, 243079, "DPX-MAT28 Technical: Subchronic Toxicity, 90-Day Feeding Study in Rats", (S. S. Anand, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.:

DuPont-21490, 2 July 2008). 15 Crl:CD(SD) rats per sex per group received DPX-MAT28 Technical (92.2%) in the diet at 0 (basal diet), 600, 2000, 6000, and 18,000 ppm for 94 to 97 days. Group mean daily intake of DPX-MAT28 Technical was 35, 114, 349, and 1045 mg/kg/day in males and 45, 146, 448, and 1425 mg/kg/day in females at 600, 2000, 6000, and 18,000 ppm, respectively. Significant treatment-related reductions in group mean bodyweight (9% (males) and 8% (females, ns) lower than controls on day 91) and bodyweight gain (15% (males) and 18% (females) lower than controls for days 0-91) were recorded at 18,000 ppm. Treatment-related changes in food consumption were not indicated. Significant reductions in food efficiency (g bodyweight gain/g food consumed) were noted for males (89% of control value) and females (83% of control value) at 18,000 ppm for test days 0-91. No treatment-related mortality, clinical signs, ophthalmology, functional observational battery and motor activity effects, hematology, serum chemistry, urinalysis, organ weight changes, gross pathology, histology, or neuropathology was indicated. Systemic NOEL = 6000 ppm (349 and 448 mg/kg/day for males and females, respectively) based reduced bodyweight, bodyweight gain and food efficiency. Neurotoxicity NOEL = 18,000 ppm (1045 and 1425 mg/kg/day for males and females, respectively). No adverse effect. Acceptable. (Green, 3/17/09)

53081-0069 243077. This position paper proposes a waiver for acute neurotoxicity testing of DPX-MAT28 Technical based on low systemic toxicity, absence of clinical observations of neurotoxicity across multiple studies, no central and peripheral nervous system involvement following 28-day and 90-day dietary exposures, and equivalence of KJM44 and MAT28 (since exposure to KJM44, the methyl ester of MAT28, is followed by rapid conversion to MAT28 by plasma esterase enzymes, the administration of KJM44 to animal models results in systemic exposures to MAT28. Both MAT28 and KJM44 have comparable pharmacokinetic profiles with the terminal metabolite for both being MAT28). (Green 4/1/09).

53081-0185; 253838; "Oral (Gavage) Acute Neurotoxicity Study of DPX-MAT28-009 in Rats"; (J.F. Barnett Jr.; Charles River Laboratories, Preclinical Services, Horsham, PA; Study No. Dupont-22792; 1/23/09); Ten Crl:CD (SD) rats/sex/group were dosed orally by gavage with 0 (vehicle: 0.5% (w/v) aqueous methylcellulose), 400, 1000 or 2000 mg/kg of DPX-MAT28-009 (Aminocyclopyrachlor technical); lot no. 009; purity: 92.2%). There were no treatment-related clinical signs or effect on mean body weights. The treatment did not affect any of the FOB parameters or motor activity assessments. The neuropathological evaluation did not reveal any treatment-related lesions. **No adverse effects evident. Reported Rat Acute Neurotoxicity NOEL:** (M/F) 2000 mg/kg (based upon the lack of treatment related effects on the 2000 mg/kg treatment group); **Study unacceptable**, possibly upgradeable to acceptable with the submission of concurrent positive control data). (Moore, 7/6/10)

METABOLISM

**53081-0087 243096, "^{14}C-DPX-MAT28: Plasma Pharmacokinetics and Pilot Material Balance in Male and Female Rats", (M.W. Himmelstein, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-22033, 18 August 2008). 1, 3, or 4 Crl:CD(SD) rats per sex per group received a single oral gavage dose of [Pyrimidine-2- ^{14}C]DPX-MAT28 at 25 or 500 mg/kg. An untreated control group (1 per sex) was used for plasma samples in the metabolism phase.

Pilot Material Balance Assay

One rat per sex received a single oral gavage dose of ^{14}C-DPX-MAT28 at 25 mg/kg. Animals were placed in glass metabolism units for collection of ^{14}C exhaled volatiles and $^{14}CO_2$ for 48 hours (at 24-hour intervals). Urine and feces were also collected at 24-hour intervals over dry ice for 168 hours. Urine, feces, tissues, carcass, cage wash, and residual feed were analyzed (LSC, HPLC, mass spectrometry (MS)) to determine preliminary material balance and tissue distribution (urine and feces were analyzed for metabolites). Radioactivity was not detected in respired breath during 48 hours after a single 25 mg/kg dose. Excretion of radioactivity occurred in both urine and feces with the majority eliminated during the first 24 hours after dosing. During 168 hours post-treatment, 36.3% (male) and 55.8% (female) of administered dose was excreted in

urine and 32.1% (male) and 51.7% (female) was eliminated in feces (24 hour values were 35.9% (male) and 53.7% (female) in urine and 31.1% (male) and 48.0% (female) in feces). The overall material balance was 69.9% (male) and 107.9% (female) after 168 hours. ^{14}C residues in tissues were generally below the limit of quantification except the male carcass contained 0.039% of administered dose. DPX-MAT28 was the only component identified in urine (35.9% (males) and 53.7% (females) of administered dose) and feces (31.1% (males) and 48.0% (females) of dose).

Plasma Pharmacokinetics Assay

4 rats per sex per group with surgically implanted jugular vein cannulas received a single oral gavage dose of ^{14}C-DPX-MAT28 at 25 and 500 mg/kg and were placed in metabolism cages. Serial blood samples were collected pre-treatment and at 5, 15, and 30 minutes, and 1, 2, 4, 8, 12, 24, and 30 hours post-dosing from the jugular vein cannula. Plasma contained quantifiable concentrations of ^{14}C residues for up to 30 hours post-treatment. Group mean plasma half-lives ($T_{1/2}$) were nearly identical for males (5.6 hours) and females (5.7 hours) at both 25 and 500 mg/kg. Group mean peak plasma concentrations (Cmax) were 3.8 μg.equiv/g for males and 5.0 μg.equiv/g for females at 25 mg/kg and 57.3 μg.equiv/g for males and 61.6 μg.equiv/g for females at 500 mg/kg (a 12 to 15 fold increase in Cmax with the 20-fold increase in dose). Times to peak plasma concentrations (Tmax) were 0.5 hours (males) and 0.4 hours (females) at 25 mg/kg and 0.6 hours (males) to 1.0 hours (females) at 500 mg/kg. Group mean area under the curve values (AUCINF) for plasma were 7.0 hr*μg/g for males and 9.0 hr*μg/g for females at 25 mg/kg and 151 hr*μg/g for males and 168 hr*μg/g for females at 500 mg/kg (21.5 (males) and 18 (females) fold increases over the low dose values). Group mean Cmax values for red blood cells were 1.3 μg.equiv/g for males and 2.0 μg.equiv/g for females at 25 mg/kg and 27.2 μg.equiv/g for males and 28.7 μg.equiv/g for females at 500 mg/kg. Group mean Tmax values for RBCs were 0.5 hour (males) and 0.3 hour (females) at 25 mg/kg and 0.6 hour for males and 1.0 hour for females at the high dose.

Plasma Metabolite Profile

3 rats per sex received a single oral gavage dose of ^{14}C-DPX-MAT28 at 500 mg/kg. Rats were sacrificed 30 minutes post-treatment and whole blood was collected and separated into plasma and erythrocytes. Additionally, 1 rat per sex was used for collection of control plasma. The only identifiable component in plasma was DPX-MAT28 at group mean concentrations of 219 μM for males and 251 μM for females. No biotransformation was indicated. DPX-MAT28 circulated in plasma and was excreted in the urine and feces as intact parent chemical. Acceptable. (Green, 3/25/09).

53146-0004; 259382; "^{14}C-Aminocyclopyrachlor (DPA-MAT28): Absorption, Distribution, Metabolism, and Elimination in the Sprague-Dawley Rat"; (M.W. Himmelstein; E.I. duPont de Nemours and Co., DuPont Haskell Global Centers for Health & Environmental Sciences, Newark, DE; Project ID No. DuPont-27389; 8/3/10); Various cohorts of Sprague-Dawley rats of both sexes were dosed orally by gavage with 25 or 500 mg/kg of [Pyrimidine-2-^{14}C] Aminocyclopyrachlor (DPX-MAT28) (batch no. 2420CEO002-1, specific activity: 42.42 uCi/mg, radiochemical purity: 99.5%) and material balance, bile duct cannulation and tissue distribution studies were performed. A multiple dosing regimen was performed in which rats of both sexes were dosed orally with 25 mg/kg/day of the radiolabeled test material. Unlabeled DPX-MAT28-012 (batch no. D100095-073, purity: 98.0%) was used to adjust the specific activity of the dosing preparations. In the material balance study, excretion in the urine constituted 40.4 to 56.5% of the administered dose as compared to 39.5 to 54.8% of the administered dose which was recovered in the feces. There was no apparent difference between the sexes or the two dose levels. Ninety to 93% of the administered dose was excreted within 24 hours for the 25 mg/kg treatment group, diminishing to 82 to 86% for the 500 mg/kg treatment group. In the bile-duct cannulation study, only a minimal amount of the administered dose was recovered in the bile, 0.13 to 0.25%. Absorption ranged from 50 to 60% of the administered dose for the 25 mg/kg treatment group and from 42 to 46% for the 500 mg/kg group (based on the data derived in the material balance study). In the tissue distribution studies, the radiolabel was largely sequestered in the GI tract and the GI contents at 1 and 6 hours post-dose. By 72 hours post-dose, the presence of

radiolabel had largely disappeared from the tissues. At any time during the tissue distribution evaluation, the presence of radiolabel in the liver was negligible. In the multiple dose regimen, within 3 to 4 days after the initial dose, an equilibrium between the urinary and fecal excretion of the radiolabel was established such that 37 to 39% of the accumulating administered dose was excreted in the urine and 55 to 57% of the dose was recovered in the feces. The test material was largely unmetabolized constituting 94 to 100% of the radiolabel which was recovered. **Study supplemental** (characterization of the pharmacokinetics was not included in the report**).** (Moore, 6/28/11)

**53081-0088 243097, "^{14}C-DPX-KJM44: Plasma Pharmacokinetics and Pilot Material Balance in Male and Female Rats", (M.W. Himmelstein, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-22375, 18 August 2008). 1, 3, or 4 Crl:CD(SD) rats per sex per group received a single oral gavage dose of [Pyrimidine-2- ^{14}C]DPX-KJM44 at 25 or 500 mg/kg. An untreated control group (1 per sex) was used for plasma samples in the metabolism phase.

Pilot Material Balance Assay

1 male and 1 female rat received a single oral gavage dose of ^{14}C-DPX-KJM44 at 25 mg/kg. Animals were placed in glass metabolism units for collection of ^{14}C exhaled volatiles and $^{14}CO_2$ for 48 hours (at 24-hour intervals). Urine and feces were also collected at 24-hour intervals over dry ice for 168 hours. Radioactivity was quantified in urine, feces, tissues, carcass, cage wash, and residual feed and urine and feces were also analyzed for metabolites (LSC, HPLC, mass spectrometry (MS)). Radioactivity was not detected in respired breath during 48 hours after a single 25 mg/kg dose. Urine was the main route of excretion of radioactivity with 80.4% (male) and 79.3% (female) of administered dose detected during 168 hours post-dosing (most (78.8% for both sexes) was excreted during the first 24 hours after dosing). 6.67% (male) and 3.06% (female) of administered radioactivity was found in feces during 168 hours with the majority excreted during 48 hours post-treatment (5.6% (male) and 2.8% (female)). Radioactivity detected in cage wash and residual food was 2.0% and 0.1% for the male and female, respectively. The overall material balance was 89.2% (male) and 82.6% (female) after 168 hours. ^{14}C residues in tissues were generally below the limit of quantification except male and female carcasses contained 0.144% and 0.121% of administered dose, respectively, and the male gastrointestinal tract and contents contained 0.009% of dose combined. DPX-MAT28 was the only metabolite identified in urine (78.8% of administered dose for both sexes) and feces (4.57% (males) and 1.72% (females) of dose).

Pharmacokinetics Assay

4 rats per sex per group with surgically implanted jugular vein cannulas received a single oral gavage dose of ^{14}C-DPX-KJM44 at 25 and 500 mg/kg and were placed in metabolism cages. Serial blood samples were collected pre-treatment and at 5, 15, and 30 minutes, and 1, 2, 4, 8, 12, 24, and 30 hours post-dosing from the jugular vein cannula. Plasma contained quantifiable concentrations of ^{14}C residues for up to 30 hours post-treatment. Group mean plasma half-lives ($T_{1/2}$) were 13.3 and 8.7 hours for males and 10.9 and 11.6 hours for females after 25 and 500 mg/kg, respectively. Group mean peak plasma concentrations (Cmax) were 20.0 μg.equiv/g for males and 16.7 μg.equiv/g for females at 25 mg/kg and 126.0 μg.equiv/g for males and 113.9 μg.equiv/g for females at 500 mg/kg (a 6 to 7 fold increase in Cmax with the 20-fold increase in dose). Times to peak plasma concentrations (Tmax) were 0.3 hours at 25 mg/kg and 0.4 to 0.5 hours at 500 mg/kg for both sexes. Group mean area under the curve values (AUCINF) for plasma were 14.8 hr*μg/g for males and 14.5 hr*μg/g for females at 25 mg/kg and 255.8 hr*μg/g for males and 241.5 hr*μg/g for females at 500 mg/kg (17.2 (males) and 16.6 (females) fold increases over the low dose values, respectively). Group mean Cmax values for red blood cells (RBCs) were 7.2 μg.equiv/g for males and 8.2 μg.equiv/g for females at 25 mg/kg and 54.7 μg.equiv/g for males and 48.1 μg.equiv/g for females at 500 mg/kg. Group mean Tmax values for RBCs were 0.3 hour for both sexes at 25 mg/kg and 0.4 hour for males and 0.6 hour for females at the high dose.

Plasma Metabolite Profile

3 rats per sex received a single oral gavage dose of ^{14}C-DPX-KJM44 at 500 mg/kg. Rats were

sacrificed 30 minutes post-treatment and whole blood was collected and separated into plasma and erythrocytes. Additionally, 1 rat per sex was used for collection of control plasma. The only identifiable component/metabolite in plasma was DPX-MAT28 at group mean concentrations of 777 μM for males and 773 μM for females. The proposed metabolic pathway (a schematic is provided) is rapid ester cleavage upon first pass metabolism in the liver and/or systemic circulation. Although not identified under conditions of this study, the metabolite IN-LXT69 (identified in a 90-day feeding study in rats, (volume 53081-0073, record 243081)), included in the pathway, is proposed to form by de-carboxylation of DPX-MA28 upon repeated daily intake of DPX-KJM44. Acceptable. (Green, 3/24/09).

SUBCHRONIC TOXICITY STUDIES

DPX-MAT 28 Rat Subchronic Dietary Toxicity Study

**53081-0070, 071 243078, 243079, "DPX-MAT28 Technical: Subchronic Toxicity, 90-Day Feeding Study in Rats", (S. S. Anand, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-21490, 2 July 2008). 15 Crl:CD(SD) rats per sex per group received DPX-MAT28 Technical (92.2%) in the diet at 0 (basal diet), 600, 2000, 6000, and 18,000 ppm for 94 to 97 days. Group mean daily intake of DPX-MAT28 Technical was 35, 114, 349, and 1045 mg/kg/day in males and 45, 146, 448, and 1425 mg/kg/day in females at 600, 2000, 6000, and 18,000 ppm, respectively. Significant treatment-related reductions in group mean bodyweight (9% (males) and 8% (females, ns) lower than controls on day 91) and bodyweight gain (15% (males) and 18% (females) lower than controls for days 0-91) were recorded at 18,000 ppm. Treatment-related changes in food consumption were not indicated. Significant reductions in food efficiency (g bodyweight gain/g food consumed) were noted for males (89% of control value) and females (83% of control value) at 18,000 ppm for test days 0-91. No treatment-related mortality, clinical signs, ophthalmology, functional observational battery and motor activity effects, hematology, serum chemistry, urinalysis, organ weight changes, gross pathology, histology, or neuropathology was indicated. Systemic NOEL = 6000 ppm (349 and 448 mg/kg/day for males and females, respectively) based reduced bodyweight, bodyweight gain and food efficiency. Neurotoxicity NOEL = 18,000 ppm (1045 and 1425 mg/kg/day for males and females, respectively). No adverse effect. Acceptable. (Green, 3/17/09).

DPX-KJM44 Rat Subchronic Dietary Toxicity Study

**53081-0072 243080, "DPX-KJM44 Technical: Subchronic Toxicity, 90-Day Feeding Study in Rats", (S.S. Anand, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-22570, 2 July 2008). 10 Crl:CD(SD) rats per sex per group received DPX-KJM44 Technical (95% aminocyclopyrachlor methyl by analysis and 89.14% DPX-MAT28 acid equivalents on a calculated basis) in the diet at 0 (basal diet), 600, 2000, 6000, and 18,000 ppm for 94 to 96 days. Group mean intake of DPX-KJM44 was 35, 117, 347, and 1022 mg/kg/day in males and 43, 133, 405, and 1219 mg/kg/day in females at 600, 2000, 6000, and 18,000 ppm, respectively. At 18,000 ppm, reductions in group mean bodyweight (8% for both sexes) and bodyweight gain (14% for males and 24% for females) were noted for treatment days 0 - 91 compared to controls (bodyweight gain decreases were statistically significant). At 6000 ppm, female final bodyweight was 6% lower than control and overall bodyweight gains (test day 0 to 91) were 15% lower vs controls (not significant). Food consumption was significantly reduced (9% for males and 11% for females) at 18,000 ppm and 9% for females at 6000 ppm relative to controls for test days 0 to 91. Overall food efficiency (g bodyweight gain/g food consumed) was 95% (males) and 85% (females) of control values at 18,000 ppm and 93% of the control value for females at 6000 ppm on test days 0 to 91. At treatment day 87, DPX-MAT28 was the predominant plasma component in the treated groups (254 and 376 ng/ml for males and females, respectively at 600 ppm and 20,660 and 29784 ng/ml for males and females, respectively at 18,000 ppm). IN-LXT69 was present in plasma at lower values at all treated levels (12.28 ng/ml in males and 15.04 ng/ml in females at 600 ppm and 438.8 ng/ml in males and 415.4 ng/ml in females at 18,000 ppm). DPX-KJM44 was below the limit of quantitation (LOQ, 10 ng/ml) in all dose groups. No treatment-

related mortality, clinical signs, ophthalmology, hematology, urinalysis, organ weights, gross pathology, or histology. Systemic NOEL = 6000 ppm (347 mg/kg/day (males) and 405 mg/kg/day (females)) based on reduced bodyweight, bodyweight gain, and food consumption. No adverse effect. Acceptable. (Green, 3/19/09).

DPX-MAT28 Mouse Subchronic Dietary Toxicity Study

**53081-0074, 0075 243082, 243083, "DPX-MAT28 Technical: Subchronic Toxicity 90-Day Feeding Study in Mice", (S.S. Anand, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-21491, 2 July 2008). 15 Crl:CD1(ICR) mice per sex per group received DPX-MAT28 Technical in the diet at 0 (basal diet), 300, 1000, 3000, and 7000 ppm for 95 to 97 days. Overall group mean daily intake of DPX-MAT28 was 47, 154, 459, and 1088 mg/kg/day for males and 67, 230, 649, and 1623 mg/kg/day in females at 300, 1000, 3000, and 7000 ppm, respectively. No treatment-related effects were noted for mortality, clinical signs, bodyweight, bodyweight gain, food consumption, food efficiency, ophthalmology, hematology, serum chemistry (total protein), organ weights, gross pathology, or histology. At treatment day 60, metabolite IN-LXT69 was below the limit of quantitation (LOQ, 10 ng/ml) in all dose groups (5 mice per sex per group were sampled) and the average concentration of DPX-MAT28 in plasma was slightly higher in males than in females at all dose levels. Values were 157 and 121 ng/ml for males and females, respectively at 300 ppm and 2484 and 2313 ng/ml for males and females, respectively at 7000 ppm. Systemic NOEL = 7000 ppm (1088 mg/kg/day (males) and 1623 mg/kg/day (females)). No adverse effect. Supplemental data. (Green, 3/20/09).

DPX-MAT28 Dog Subchronic Dietary Toxicity Study

**53081-0076, 0077 243084, 246476, 243085, "DPX-MAT28 Technical: Subchronic Toxicity 90-Day Feeding Study in Dogs, Revised Final Report", (E.M. Luckett, MPI Research, Inc., Mattawan, MI, MPI Research Study No. 125-068, 2 July 2008). 4 Beagle dogs per sex per group received DPX-MAT28 Technical (92.2% aminocyclopyrachlor) in the diet at 0 (basal diet), 250, 1250, 5000, and 15000 ppm for 91 days. Overall mean daily intake values of DPX-MAT28 Technical were 6.46, 33.31, 126.23, and 425.71 mg/kg/day for males, and, 7.02, 37.94, 124.12, and 387.53 mg/kg/day for females at 250, 1250, 5000, and 15000 ppm, respectively. All animals survived to terminal necropsy. Treated males and females were observed with scaling skin on the feet (forepaws and hindpaws). All treated groups were affected and, while the high dose group had the highest incidence, a dose response pattern was not indicated. Scaling was not present in control animals. No treatment-related neurobehavioral findings were noted. Food consumption, bodyweight, hematology, serum chemistry, necropsy, and histology were not affected by treatment. Blood samples were collected from all animals (not fasted) once during week 9 for determination of plasma concentrations of DPX-MAT28 Technical and metabolite IN-LXT69. Metabolite IN-LXT69 was identified in plasma at the high dose (11.7 and 11.6 ng/ml in males and females, respectively) and was below the limit of detection (10 ng/ml) at all other dose levels. DPX-MAT28 Technical was the main analyte and levels in plasma were higher for females vs males at all dose levels. High dose values for DPX-MAT28 Technical were 63000 ng/ml (males) and 82575 ng/ml (females). At terminal necropsy, a sample of liver was collected from each animal and analyzed for total cytochrome P-450 and for quantification of cytochrome P-450 1A1, 1A2, 2B1/2, 2E1, 3A2, and 4A1/2/3. In males at 5000 and 15000 ppm, increases in cytochrome P-450 1A1 (501% and 518% of control, respectively) and 2B1/2 (225% and 191% of control, respectively) were noted (values at 5000 ppm were statistically significant). Additionally, in males at 15000 ppm, a statistically significant decrease in P-450 4A1/2/3 was noted (89% of control). In females, increases in cytochrome P-450 2B1/2 (193% and 134% of control at 5000 and 15000 ppm, respectively (statistically significant at 5000 ppm only)) and in P-450 4A1/2/3 (113%, 109%, and 115% of control at 1250, 5000, and 15000 ppm, respectively (increases at 1250 and 15000 ppm were statistically significant)) were noted. Additionally, P-450 2E1 was significantly increased in females at 1250 ppm (187% of control). NOEL = 15000 ppm (425.71 mg/kg/day in males and 387.53 mg/kg/day in females). No adverse effect. Acceptable. (Green, 4/15/09).

DPX-MAT28 Rat 28-Day Repeated Dose Dermal Toxicity Study

**53081-0078 243086, "DPX-MAT28 Technical: Dermal Toxicity Study (28 Day Repeat Dermal Application Study in Rats)", (D.J. Merkel, Eurofins, Product Safety Laboratories, Dayton, OH., Laboratory Project ID: DuPont-22796, 28 March 2008). 10 Hsd:Sprague Dawley® rats per sex per group were dermally treated (clipped, unabraded, occluded) with DPX-MAT28 Technical (90.9%) at 0 (distilled water), 150, 400, and 1000 mg/kg/day for 28 days (6 hours per day, 7 consecutive days per week). No treatment-related effects were indicated for mortality, clinical signs of toxicity, ophthalmology, bodyweights, food consumption, hematology, serum chemistry, necropsy results, or histology at any treatment level. Very slight erythema was noted on the treated skin of one high dose male on day 3 and of one 400 mg/kg/day male and one high dose male on day 28. No other test substance-related dermal irritation (erythema or edema) was noted during the study. Systemic NOEL = 1,000 mg/kg/day. No adverse effect. Acceptable. (Green, 3/11/09).

DPX-MAT28 Mouse 28-Day Immunotoxicity Studies

**53081-0089 243098, "DPX-MAT28 Technical: 28-Day Immunotoxicity Feeding Study in Male Rats", (D. Hoban, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-22794, 18 June 2008). 10 Crl:CD(SD) male rats per group received DPX-MAT28 Technical (92.2%) in the diet at 0 (basal diet), 600, 6000, 18,000 ppm for 28 days. All animals were injected intravenously (IV) (lateral tail vein) with 0.5 ml of 4 x 10^8 sheep red blood cells (sRBC)/ml on test day 22. Animals were sacrificed on test day 28 and 4 ml of blood were collected (abdominal *vena cava*) from each animal for humoral immune function. Sera samples were evaluated using sRBC specific IgM levels with an enzyme-linked immunosorbent assay (ELISA). The overall group mean daily intake of DPX-MAT28 was 42, 407, and 1277 mg/kg/day at 600, 6000, and 18,000 ppm, respectively. No treatment-related effects were noted for clinical signs, bodyweight, bodyweight gain, food consumption, gross pathology, organ weights, or the primary humoral immune response. The positive control was functional. No immunotoxicity, no adverse effects. Clinical and Immuntoxicity NOEL = 18,000 ppm (1277 mg/kg/day). Acceptable. (Green 3/26/09).

**53081 243099, "DPX-MAT28 Technical: 28-Day Immunotoxicity Feeding Study in Male Mice", (D. Hoban, E.I. DuPont de Nemours and Company, DuPont Haskell Global Centers for Health and Environmental Sciences, Newark, DE., Laboratory Project ID.: DuPont-22795, 18 June 2008). 10 Crl:CD1(ICR) male mice per group received DPX-MAT28 Technical (92.2%) in the diet at 0, 300, 3000, and 7000 ppm for 28 days. On test day 23, all animals were injected intravenously (IV) (lateral tail vein) with 0.2 ml of 5 x 10^8 sheep red blood cells (sRBC)/ml. Animals were sacrificed on test day 28 and 1 ml of blood was collected (abdominal *vena cava*) from each animal for humoral immune function. Sera samples (including those from the positive control animals) were evaluated using sRBC specific IgM levels with an enzyme-linked immunosorbent assay (ELISA). The overall group mean daily intake of DPX-MAT28 was 45, 425, and 1056 mg/kg/day at 300, 3000, and 7000 ppm, respectively. All mice survived to scheduled sacrifice. No treatment-related effects were noted for clinical signs, bodyweight, bodyweight gain, food consumption, gross pathology, organ weights, or the primary humoral immune response. The positive control was functional. No immunotoxicity, no adverse effects. Clinical and Immunotoxicity NOEL = 7000 ppm (1056 mg/kg/day). Acceptable. (Green, 3/27/09).