

CALIFORNIA ENVIRONMENTAL PROTECTION AGENCY
DEPARTMENT OF PESTICIDE REGULATION
MEDICAL TOXICOLOGY BRANCH

SUMMARY OF TOXICOLOGY DATA

METHAMIDOPHOS

Chemical Code # 1697, Tolerance # 315
SB 950 # 2

July 20, 1993

Revised: 6/16/94, 5/30/95, 2/6/96, 8/20/97, 6/22/98, 11/22/99, 9/30/02, 10/01/03 and 6/18/04

I. DATA GAP STATUS

Chronic toxicity, rat:	No data gap, possible adverse effect
Chronic toxicity, dog:	No data gap, possible adverse effect
Oncogenicity, rat:	No data gap, no adverse effect
Oncogenicity, mouse:	No data gap, possible adverse effect
Reproduction, rat:	No data gap, possible adverse effect
Teratology, rat:	No data gap, possible adverse effect indicated
Teratology, rabbit:	No data gap, no adverse effect
Gene mutation:	No data gap, possible adverse effect
Chromosome effects:	No data gap, possible adverse effect
DNA damage:	No data gap, no adverse effect
Neurotoxicity ¹ :	No data gap, no adverse effect

Toxicology one-liners are attached.

All record numbers through 211501 and 960217 were examined.

** indicates an acceptable study.

Bold face indicates a possible adverse effect.

File name: T040618.wpd

Revised by S. Morris, 11/22/99 and Gee, 9/30/02, 10/01/03 and 6/18/04

¹ Rat neurotoxicity studies are on file. A developmental neurotoxicity study is on file.

Note: 315-122; 089116; "SRA 5172 Study of the Subchronic Inhalation Toxicity to Rats in Accordance with OECD Guideline No. 413", Laboratory Project ID Report No. 98370; J. Pauluhn, Bayer AG, Wuppertal, Germany; 3/30/88. This study was not a required test type and was therefore not evaluated for acceptability and no worksheet was done (S. Morris, 1/5/93).

Note: 315-120; 089114: Supplemental data for doc. # 315-122, rec. # 089116. In the course of reviewing the recent genotoxicity studies and preparing the Summary of Toxicology Data, older studies were rereviewed. A number were upgraded as noted in the one-liners (S. Morris, 7/20/93).

II. TOXICOLOGY ONE-LINERS AND CONCLUSIONS

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

COMBINED, RAT

**** 315-060; 019914;** "Chronic Feeding/Oncogenicity Study of Technical methamidophos (Monitor®) to Rats", Study No. 81-271-01, Mobay No. 88687; R.H. Hayes, Mobay Chemical Corporation, Stilwell, KS, 11/13/84. Technical Methamidophos (Monitor, 70% stated purity, batch # 77-297-149) was fed in the diet to groups of 60 Fisher 344 rats/sex/dose for 106 weeks at 0, 2, 6, 18, or 54 ppm. Ten rats/sex/dose were sacrificed at 52 weeks. There were no treatment-related oncogenic effects. Treatment-related effects were: reduced group body weights in both sexes at 54 ppm with group mean body weights always being > 84% of controls and reduced group mean relative testicle weights in males at 18 and 54 ppm. Cholinesterase levels in brain, erythrocyte, and plasma were reduced in both sexes at 2, 6, 18, and 54 ppm (NOEL < 2 ppm). A **possible adverse effect** was indicated by approximately 10, 35, 64, and 77% depression of brain cholinesterase activity at respectively 2, 6, 18, and 54 ppm. The study was unacceptable (J. Schreider 1/29/85; H. Green and S. Morris, 11/17/92) but upgraded to acceptable by submission of adequate analytical data, individual clinical data, and rationale for the doses used (S. Morris, G. Patterson, and J. Gee; 5/30/95) .

315-065; 027092: Partial duplicate of doc. # 315-060, rec. # 019914.

315-106; 075900: This document contains gross and histopathology data for rats sacrificed at 12 months and historical control data for organ weights in Fisher 344 rats.

315-137; 130534: This document contains adequate analytical and individual clinical data (S. Morris, 5/30/95).

315-137; 130552: This document contains a 5-week pilot study in which 5 Fischer 344 rats/sex/dose were fed diets containing 0, 1, 2, 4, 8, 16, 32, and 64 ppm. There were no treatment-related clinical signs or pathological findings. There appeared to be a difference in group mean male body weights from controls at 64 ppm. The significance of this difference was obscured by the low number of animals and high variability within and between treatment groups. There were dose-related decreases in plasma, erythrocyte, and brain cholinesterase levels when compared to controls. Brain cholinesterase levels were not measured for the 32 and 64 ppm treatment groups. Evaluation of these data resulted in a change of study status to acceptable (S. Morris, G.

Patterson, and J. Gee; 5/30/95).

Note: The possible adverse effect was listed under the Chronic Toxicity test type for rat.

CHRONIC TOXICITY, RAT

See COMBINED, RAT above.

315-013; 960209; "Two-year Chronic Oral Toxicity of RE 9006-III, SX-116 in Albino Rats", IBT No. B5485, 2/18/70; invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 10/16/92).

CHRONIC TOXICITY, DOG

**** 315-061; 019916;** "One-Year Feeding Study of Methamidophos (Monitor®) in Dogs", R. H. Hayes, Mobay Chemical Corporation, Stilwell, Kansas, Study No. 81-174-01, Mobay No. 87474; 6/26/84. Technical methamidophos (Monitor, 70% stated purity, batch # 77-297-149) was fed in the diet of 6 beagle dogs/sex/group for 52 weeks at 0, 2, 8, or 32 ppm. There were no significant treatment-related effects on: clinical signs, feed consumption, body weight or ophthalmology, gross pathology, histopathology, hematology, and urology findings (chronic NOEL \geq 32 ppm). Plasma, erythrocyte and brain cholinesterase activity were depressed at all doses. A **possible adverse effect** was indicated by a treatment-related depression of brain cholinesterase activity which was approximately 85, 50, and 30% of controls at respectively 2, 8, and 32 ppm (NOAEL = NOEL < 2 ppm). The study was unacceptable (J. Schreider, 1/25/85; S. Morris and H. Green, 10/15/92) but upgraded to acceptable by submission of adequate analytical data and rationale for the doses used (S. Morris, G. Patterson, and J. Gee; 5/30/95).

315-065; 027093: Partial duplicate of doc. # 315-061, rec. # 019916.

315-107; 075901: This document contains ophthalmology data for doc. # 315-061, rec. # 019916.

315-138; 130795: This document contains adequate analytical data and rationale for the doses used. Evaluation of this submission resulted in a change of study status to acceptable (S. Morris, G. Patterson, and J. Gee; 5/30/95).

315-013; 960208: "Two-year Chronic Oral Toxicity of RE9006-III, SX-116 in Beagle Dogs", IBT No. C5486, 10/20/69; invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 10/15/92).

ONCOGENICITY, RAT

See COMBINED, RAT above.

ONCOGENICITY, MOUSE

**** 315-059; 019913;** "Oncogenicity Study of Methamidophos Technical (Monitor®) on Mice." (Study No. 80-332-01, Mobay No. 87479, R. H. Hayes, Mobay Chemical Corporation, Stilwell, KS, 8/6/84). Technical methamidophos (Monitor, 70% stated purity, batch # 77-297-149) was fed in the diet of 60 CD1 albino mice/sex/group for 106 weeks at 0, 1, 5, or 25 ppm. Ten mice/sex/group were sacrificed at week 53. There were no treatment-related effects on survival or tumor incidence. Treatment-related effects were decreased body weight gains in both sexes after one year at 25 ppm with all treatment groups have group mean body weights > 86% of controls. A dose-related increase in diffuse interstitial pneumonia was seen in both sexes at 1, 5, and 25 ppm but not noted as a possible adverse effect because it was probably secondary to poor husbandry. **A possible adverse effect** was indicated by a treatment-related depression of brain cholinesterase activity in a pilot study (DPR doc. # 315-136, rec. # 130533). Compared to controls, brain cholinesterase activity was depressed approximately 25, 60, 87, and 90% at respectively 2, 10, 50, and 100 ppm. No evidence for an oncogenic effect was seen. The study was unacceptable (J. Schreider, 1/29/85; S. Morris and H. Green, 10/27/92) but upgraded to acceptable with submission of an adequate rationale for the doses used and adequate analytical data (S. Morris, G. Patterson, and J. Gee; 5/30/95).

315-065; 027094: Partial duplicate of doc. # 315-059, rec. # 019913.

315-107; 075902: Histopathology data for the 10 mice/sex/group that were sacrificed at week 53.

315-136; 130533: This document contains adequate analytical data (S. Morris, DPR Response, 1/12/95).

315-136; 130549: This document contains a 6-week pilot study in which at least 20 CD1 albino mice/sex/dose were fed diets containing 0, 2, 10, 50, and 100 ppm. There was 15% decrease in group mean body weight in males in the first week of treatment at 100 ppm. This was followed by normal body weight gain throughout the rest of the study. Depression of plasma, erythrocyte, and brain cholinesterase levels were treatment-related. Other possible signs of toxicity were seen at 100 ppm: two males had wet abdomens and one female had a black tarry-like substance around the anal region. Evaluation of these data resulted in a change of study status to acceptable (S. Morris, G. Patterson, and J. Gee; 5/30/95).

REPRODUCTION, RAT

****315-157; 159401;** "A Two-Generation Dietary Reproduction Study in Rats Using Technical Methamidophos." (Study No. 95-672-GJ; D.A. Eigenberg, K.J. Freshwater, and S.G. Lake; Bayer Corporation, Stilwell, KS; 1/5/98.) Groups of 30 Sprague-Dawley rats/sex were fed methamidophos (69.0% to 76.7% purity) in the diet at nominal concentrations of 0, 1.0, 10.0, and 30.0 ppm (analytical concentrations were 0, 1.0, 9.7, 26.1 ppm). Exposures were continuous for the P and F1 adults. Starting at 7 weeks of age P adults were exposed for 10 weeks then mated twice to produce the F1a and F1b litters. Randomly-selected F1b weanling pups were exposed

for 10 weeks then mated twice to produce the F2a and F2b litters. F1a, F2a, F2b, and unselected F1b pups were sacrificed at weaning. P and F1 males were sacrificed after the second breeding of each generation. Females were sacrificed at the respective F1b and F2b weanings. Adult animals were evaluated for compound-related effects on body weight, food consumption, clinical signs, estrous cycling, mating fertility, gestation length, litter size, and cholinesterase levels. The offspring were evaluated for effects on sex ratios, pup viability, body weight gain, clinical signs, cholinesterase activity. Gross necropsy evaluations were performed on all adults and pups. Histopathologic evaluations of the reproductive organs, the pituitary, and gross lesions were performed on all P and F1 adults. Treatment-related parental effects included: decreased body weight gain in P (30.0 ppm) and F1 (10.0, 30.0 ppm) males, F1 females (10.0, 30.0 ppm), and lactating females (10.0, 30.0 ppm); increased food consumption in P and F1 males (30.0 ppm); and decreased food consumption in lactating females (30.0 ppm) (parental NOEL = 1.0 ppm). A **possible adverse effect** was indicated by a treatment-related decrease in pup body weight gain seen at 10.0 and 30.0 ppm and increased pup cannibalism and decreased survival seen at 30.0 ppm (developmental NOEL = 1.0 ppm). Cholinesterase (ACh) inhibition was seen in adults in plasma, erythrocyte, and brain at 1.0, 10, and 30 ppm (adult ACh inhibition NOEL < 1.0 ppm). Cholinesterase inhibition was seen in pups in plasma, erythrocyte, and brain at 10 and 30 ppm (pup ACh inhibition NOEL = 1.0 ppm). The study is acceptable (S. Morris and J Gee, 4/9/98).

315-061; 019915; "Effect of Methamidophos (Monitor®) on Reproduction in Rats." (Study No. 82-671-01, E.J. Hixson, Mobay Chemical Corporation, Stilwell, KS, 11/8/84.) Methamidophos Technical (Monitor, 70.5% stated purity, batch # 77-297-149) was fed in the diet at nominal concentrations of 0, 3, 10, or 33 ppm to 26 CD adult rats/sex/group continuously through 2 generations beginning with at least 100 days of exposure for the adult F0 parents before mating then through gestation and lactation of a single F1 litter, during a 120-day growth phase of F1 rats and through gestation, lactation and one month rest period between mating for the F2a and F2b litters. Mean analytical concentrations were 0, 3.0, 9.1, and 29.9 ppm. There were no significant treatment-related effects on the parental animals (parental NOEL \geq 33 ppm). A **possible adverse effect** was indicated by decreased mean live litter size and pup growth at 33 ppm and decreased live litters at 3, 10 and 33 ppm (reproductive NOEL < 3 ppm). The study is unacceptable and not upgradeable because there were less than 20 litters per treatment group, male reproductive performance could not be evaluated, inadequate rationale for high dose, inadequate necropsy and histopathology data, and no reproductive NOEL (J. Schreider, 1/28/85; H. Green and S. Morris, 10/2/92).

315-065; 027100: Partial duplicate of doc. # 315-061, rec. # 019915.

315-101; 072564: This document contains a rebuttal to EPA evaluations of doc. # 315-061, rec. # 019915 and additional data.

315-013; 960211; "Three-generation Reproduction Study in Albino Rats on SX-171 (Technical RE-9006-75%), Results Through Weaning of F1b Litters (First Generation), S-90", IBT No. P6255, 2/19/69; invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 11/12/92).

315-013; 960212; "Three-generation Reproduction Study in Albino Rats - SX-171 (Technical RE-9006-75%), Results Through Weaning of F2b Litters (Second Generation), Chevron Request No. S-90", IBT No. P6255, 9/12/69; invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 11/12/92).

315-013; 960214; "Three-generation Reproduction Study in Albino Rats on SX-171 (Technical RE-9006 75%), Results of All Three Generations, Chevron Request No. S-90", IBT No. P6255, 1/16/70; invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 11/12/92).

TERATOLOGY, RAT

**315-149; 144391; "Developmental Toxicity Study with Monitor® Technical in Sprague-Dawley Rat." (Study No. 96-612-EM; Bayer Report 107178; A.B. Astroff; Bayer Corporation, Agricultural Division, Toxicology, Stilwell, KA; 1/17/96.) Monitor Technical (batch no. # 0067009, 76.0% purity) was given to 36 mated female Sprague-Dawley rats/group by oral gavage at nominal doses of 0, 0.04, 0.1, or 4.0 (analytical 0, 0.05, 0.14, or 5.49) mg/kg/day on gestation days 6 through 15. Erythrocyte, plasma, and brain cholinesterase levels were determined on 6 rats/group that were sacrificed 90 minute after the last dose on gestation day 15. The remaining 30 animals/group were sacrificed on day 20, gross necropsy were performed, intact uteri were removed and weighed, fetuses sacrificed and weighed. Reproductive indices were measured and the fetuses examined for gross external malformations. One half of the fetuses were examined for visceral abnormalities and the other half cleared, stained and examined for skeletal abnormalities. Treatment-related maternal effects seen at 5.49 mg/kg/day were: increased incidence of tremors, muscle fasciculation, and salivation; decreased food consumption and body weight gain; and decreased plasma, erythrocyte, and brain cholinesterase activities (maternal NOEL = 0.14 mg/kg/day). Treatment-related fetal effects seen at 5.49 mg/kg/day were: decreased group mean fetal weight and an increased incidence of incomplete ossification of sacral arches and sternebra (developmental NOEL = 0.14 mg/kg/day). No adverse effect was indicated. The study was unacceptable but upgraded by an adequate rationale for dose selection. See doc. # 315-059, rec. # 20042 below and DPR Response, 11/22/99; S. Morris. 11/22/99).

315-155; 149039; "A Dose Range-Finding Developmental Toxicity Study with MONITOR Technical in the Sprague-Dawley Rat," study Number 94-612-ZA; A.B. Astroff; Bayer Corporation, Agricultural Division, Toxicology Department, Stilwell, KA. Nominal doses of 0.0, 0.04, 0.1, 1.0, or 4.0 mg/kg/day (analytical 0.0, 0.05, 0.15 1.41 or 6.04 mg/kg/day) were given by oral gavage to groups of 6 pregnant Sprague-Dawley rats on gestation days 6 through 20. Treatment-related maternal effects included: increased incidences of tremors, muscle fasciculation and decreased food consumption and body weight gain at 6.04 mg/kg/day and decreased plasma and brain cholinesterase levels at 0.15, 1.41, and 6.04 mg/kg/day and erythrocyte cholinesterase levels at 1.41, and 6.04 mg/kg/day. The only treatment-related developmental effect seen was reduced group mean fetal weight at 6.04 mg/kg/day. Evaluation of these data did change the study status of DPR doc. # 315-149, rec. # 144391 (Morris, DPR Response, 6/22/98).

315-163; 169761: The registrant submitted comments about dose analysis, rationale for dose selection, and correcting the NOEL based on dose analysis. Evaluation of this submission did not result in a study status change. No worksheet was done (see DPR Response, 11/22/99; S. Morris. 11/22/99).

315-059; 20042: Acute Oral Toxicity Study; Rat; Mobay Chemical Corporation, Corporate Toxicology Department, Stanley Research Center, Stilwell, KS; Report No. 68802; 7/1/80). These data have been reviewed and found acceptable (T. Moore,

6/29/99; SRS 7/1/99). A formulated product containing 40% methamidophos had an acute oral LD50 for female rats of 21.3 mg/kg. The expected LD50 for monitor technical (76% methamidophos) would be approximately 10.8 mg/kg. The clinical signs and cholinesterase levels seen in the present study, the calculated LD50 and the expected presentation of organophosphorus toxicity indicate that the high dose (5.49 mg/kg) approached lethality. This is an adequate rationale for the dose selection in doc. # 315-149, rec. # 144391. The study is acceptable. No worksheet was done (see DPR Response, 11/22/99; S. Morris, 11/22/99).

315-061; 019917; "Embryotoxic and Teratogenic Effects of Methamidophos (Monitor®) in Rats." (Study No. 82-611-01, Mobay No. 87480, E. J. Hixson, Mobay Chemical Corporation, Stilwell, KS, 10/15/84.) Methamidophos (Monitor, 70.5% stated purity, batch # 77-297-149, water vehicle) was given by oral gavage to groups of 24 to 27 mated (sperm-positive, gestation day 0) female CD rats on gestation days 6 through 15 at 0, 0.3, 1.0, or 3.0 mg/kg/day. Sacrifice and Cesarean section were conducted on gestation day 21. Treatment-related maternal effects seen at 3.0 mg/kg were: decreased body weight gains with group mean body weights always > 88% of controls, decreased feed consumption, and increased clinical signs of cholinesterase inhibition (fasciculations, salivation, lacrimation, hyperactivity, and excessive urination; maternal NOEL = 1.0 mg/kg/day). There were no treatment-related effects on fecundity or skeletal or organ abnormalities. A **possible adverse effect** was indicated by decreased group mean fetal weights at 3.0 mg/kg in the main study and aborted litters in a preliminary study at 4.5 mg/kg/day (developmental NOEL = 1.0 mg/kg/day). The study was unacceptable but possibly upgradeable with adequate analytical data, submission of preliminary studies, and rationale for the doses used (J. Schreider, 1/29/85; H. Green and S. Morris, 11/06/92).

315-065; 027101: Partial duplicate of doc. # 315-061, rec. # 019917.

315-065; 027102: "A Pilot Teratology Study Using Technical Methamidophos in CD Rats", Study No. 80-611-01, Mobay No. 68765, Methamidophos (Monitor, 70% stated purity, batch # 77-297-149) was given by oral gavage to groups of 4 mated female rats at 0.0, 0.1, 0.3, 1.0, 3.3, or 10.0 mg/kg/day on gestation days 6 through 20. The only treatment-related effects reported were seen at 3.3 and 10 mg/kg/day: mild to severe signs of organophosphate toxicity, decreased mean maternal weight gain and decreased mean fetal birth weights. The report "recommended that the high dose level, for the definitive teratology study incorporating Methamidophos, not be higher than 10 mg/kg/day." No worksheet was done (S. Morris, 11/9/92).

TERATOLOGY, RABBIT

315-150; 145998; "Oral (Stomach Tube) Developmental Toxicity Study of MONITOR® Technical in Rabbits." (Argus # 222-001, Valent # VP-10143; A.M. Hoberman; Argus Research Laboratories, Inc., Horsham, PA; 3/8/89.) Groups of 23 New Zealand White pregnant female rabbits were given methamidophos (Monitor®, lot 0067009, 76% purity, water vehicle) by oral gavage at 0 (10 ml/kg water), 0.1, 0.5, or 2.5 (analytical 0, 0.2, 0.65, or 2.47) mg/kg/day on gestation days 6 through 18. All dams were sacrificed on gestation day 29, Caesarean-sectioned, examined grossly for internal lesions, corpora lutea counted, and uteri weighed and examined in detail. All fetuses were weighed, externally examined for alterations, and internally examined for visceral and skeletal alterations. Transient depression in body weight gain and

food consumptions were seen at 0.65 and 2.47 mg/kg/day. Hyperactivity was observed at 2.47 mg/kg/day. Maternal NOEL = 0.2 mg/kg/day. There were no treatment-related effects on gestational observations, uterine weights, litter observations, fetal observations, and fetal malformations and abnormalities. Fetal NOEL \geq 2.47 mg/kg/day. No adverse effect was indicated. The study was unacceptable and not upgradeable because the rationale for the doses used was inadequate (4/22/97, S. Morris and J. Gee).

315-150; 145998; pp. 181 - 316. Dose selection for the main study above was based on a preliminary pilot study: "Oral (Stomach Tube) Dosage-Range Developmental Toxicity Study of Monitor[®] in Rabbits," Argus # 222-001, Valent # VP-101143; A.M. Hoberman; Argus Research Laboratories, Horsham, PA, 3/8/96. Groups of 5 New Zealand White pregnant female rabbits were given methamidophos (Monitor[®], lot 0067009, 76% purity, water vehicle) by oral gavage at 0 (10 ml/kg water), 0.1, 0.5, 2.5, 5.0, or 7.5 (analytical 0, 0.2, 0.46, 2.46, 4.90, or 7.73) mg/kg/day on gestation days 6 through 18. Blood samples were taken on gestation day 18 to determine plasma (PChE) and erythrocyte (RChE) cholinesterase levels. All dams were sacrificed on gestation day 29, Caesarean-sectioned, and the uterine contents were examined. There were treatment-related maternal effects: death (2/5) at 7.73 mg/kg/day; rapid breathing, excess salivation, ataxia, abnormal breathing, and decreased food consumption at 4.90 and 7.73 mg/kg/day; decreased body weight gain at 2.46, 4.90, and 7.73 mg/kg/day; and decreased PChE and RChE levels at 0.46, 2.46, 4.90, and 7.73 mg/kg/day. The only treatment-related fetal effect was decreased group mean weight at 2.46, 4.90, and 7.73 mg/kg/day. These data did not adequately support a rationale for the doses used in the main study (4/22/97, S. Morris and J. Gee).

315-156; 159400: The registrant submitted comments about DPR's evaluation of the study at DPR doc. # 315-150, rec. # 145998; quotes of U.S. EPA policy, and two tables of cholinesterase inhibition in rats treated with methamidophos, and a table of similar data from rabbits that has already been reviewed by DPR. Evaluation of this submission did not result in a change in study status (S. Morris, DPR Response, 4/22/98).

315-162; 168533: The registrant submitted comments about DPR's evaluation of the study at DPR doc. # 315-150, rec. # 145998; and tables of data previously evaluated by DPR. Evaluation of this submission did not result in a change in study status (S. Morris, DPR Response, 11/22/99; J. Gee, DPR Meeting Memo, 2/18/99).

315-027; 001213 "SRA 5172 (Methamidophos), Studies of Embryotoxic and Teratogenic Effects on Rabbits Following Oral Administration." (Report No. 8410, L. Machemer, Bayer AG, Institut für Toxikologie, Wuppertal-Elberfeld, Germany, 5/31/79.) Methamidophos (SRA 5172, 62% stated purity, suspended in 0.5% Cremophor emulsion) was given by oral gavage to groups of 15 naturally-inseminated (gestation day 0) female Himalayan rabbits on gestation days 6 through 18 at 0, 0.1, 0.5, or 2.5 mg/kg/day. Cesarean sections and sacrifices were performed on gestation day 29. There were biologically insignificant and non-dose related reductions in doe body weight gain in all treatment groups. There were no other treatment-related maternal or fetal effects (maternal and fetal NOEL's \geq 2.5 mg/kg/day). No adverse effect was indicated. The study is unacceptable but possibly upgradeable with submission of adequate analysis of the test and dosing materials, individual maternal and fetal data, and rationales for the doses and vehicle used (J. Schreider, 1/29/89; H. Green and S. Morris, 10/29/92).

315-001; 960210: Summary of doc. # 315-027, rec. # 001213.

315-029; 001214: Exact duplicate of doc. # 315-027, rec. # 001213.

315-050; 017047: Exact duplicate of doc. # 315-027, rec. # 001213.

Summary: The collective data for rabbit teratology studies with methamidophos (DPR doc. #'s 315-150, 315-156, 315-162, 315-027; rec. #'s 145998, 159400, 168533, 001213), have been reviewed. A pilot study that used a dose-range, marginally higher than the main study, that produced treatment-related effects on pregnant rabbits that included: death (2/5) at 7.73 mg/kg/day; decreased food consumption at 4.90 and 7.73 mg/kg/day, and decreased body weight gain a 2.46, 4.90, and 7.73 mg/kg/day death. The only treatment-related effect seen on uterine, fetal, or reproductive parameters appears to be a decrease in group mean fetal weights. The collective data are adequate to fill the data gap for a rabbit teratology study (see DPR Response, 11/22/99; S. Morris and J. Gee, 11/22/99).

GENE MUTATION

315-147; 141465; "CHO/HGPRT Mutation Assay," (Study No. TC865.332, Bayer No. 105076; C.A.H. Bigger and C. I. Sigler; Microbiological Associates, Inc. Rockville, MD; 5/27/93) Forward mutation of the hypoxanthine-guanine phosphoribosyl transferase (HGPRT) locus of Chinese hamster ovary (CHO) cells was measured after exposures to Methamidophos (batch no. 0-06-7009) at 0, 0 (solvent) 1.0, 2.0, 3.0, 4.0, or 5.0 mg/ml with or without S-9 metabolic activation (9000 g supernatant of Aroclor-induced, male Sprague-Dawley rat liver homogenate). Duplicate flasks were seeded with 5×10^5 cells, grown for 18-24 hours, incubated with medium containing the test material with or without S-9 for 5 hours, washed, and re-incubated for 18-24 hours. Cytotoxicity was assessed by replating 1 replicate in triplicate at 100 cells/60 mm dish and incubating for 7-10 days. Expression of the mutant thioguanine-resistant phenotype was assessed by replating remaining replicates in duplicate and subculturing every 2-3 days for 7-9 days. Selection of the mutant phenotype was done by replating each duplicate into 5 flasks and incubating in the presence of 10 μ M 6-thioguanine (TG) for 7-10 days. Colonies were fixed, stained, and counted for cloning efficiency and mutant selection. A **possible adverse effect** was indicated by increased TG resistant colonies seen at 5.0 mg/ml with metabolic activation. The study is unacceptable and not upgradeable because there was only one trial and the concentrations were inadequate (S. Morris and J. Gee, 4/10/97).

** 315-121; 089115; "CHO/HGPRT Mutation Assay." (Laboratory Study Number T5844.332008; J. W. Harbell and D. Jacobson-Kram, Microbiological Associates Inc., Rockville, Maryland, 1/12/90.) Methamidophos (Monitor Technical, # 77-297-149, 72.9% stated purity, DMSO solvent) for the ability to induce forward mutations at the hypoxanthine-guanine phosphoribosyl transferase (HGPRT) locus of Chinese hamster ovary (CHO) cells. CHO cells were plated at 5×10^5 cells/25 cm² and incubated 18-24 hours. Duplicate flasks were incubated for 5 hours in medium containing the test material at 0.0, 0.2, 0.5, 1.0, 2.0, or 3.5 μ l/ml and with or without S-9 metabolic activation system (9,000 x g supernatant of Aroclor-1254 induced, male Fisher 344 rat liver homogenates). Replicate cells were then harvested and pooled. To assess cytotoxicity cells were plated in triplicate at 100 cells/60 mm dish. For expression, pooled cells were subcultured every 2-3 days for 7 to 10 days at 106 cells/100 mm dish and finally harvested. For selection of the thioguanine (TG)-resistant phenotype, cells were plated in 4 dishes at 2×10^5 cells/100 mm dish in medium containing 10 mM TG. For cloning efficiency at selection, cells

were plated in triplicate at 100 cells/60 mm dish. For cytotoxicity, selection, and cloning efficiency at selection the final incubation was 7 days after which colonies were fixed, stained and counted. No adverse effect was indicated by no treatment-related effect on TG-resistant colony count. The study is acceptable (J. Gee and S. Morris, 12/9/92).

315-143; 137332; "SRA 5172: Salmonella/Microsome Test," (Bayer Report No. 106392; B. Herbold; B. Bayer AG Department of Toxicology, Wuppertal, Germany; 9/14/94.) The frequency of reversion of histidine auxotrophic strains of *Salmonella typhimurium* (TA 1535, TA 1537, TA 100, and TA 98) to prototrophy was measured after exposures to methamidophos (SRA 5172, batch number 278167052, 73.4% purity, water vehicle) for 48 hours at 0, 16, 50, 158, 500, 1581, or 5000 µg per plate without or with S9 metabolic activation system (9000 g supernatant of homogenized livers from Aroclor-1254-induced male Sprague-Dawley rats). There were 4 replicates per dose per strain. Exposure time was for forty-eight hours. There were 3 trials with S9 and 2 without. Controls were adequate. No treatment related increase in reversion rate or bacteriotoxic activity was seen. No adverse effect was indicated. The study was unacceptable but possibly upgradeable with submission of adequate analysis of the exposure solutions (J. Kishiyama and S. Morris, 4/1/97).

315-061; 019920; "Salmonella/Microsome Test to Evaluate for Point Mutation." (Report No. 9175, B. Herbold, Bayer AG, Wuppertal-Elberfeld, 5/20/80.) SRA 5172 Methamidophos (SRA 5172, 62.6% stated purity, DMSO solvent) was tested in a bacterial assay that measured the rate of mutation of a histidine auxotrophic strains of *Salmonella typhimurium* (tester strains TA98, TA100, TA1535, and TA1537) to prototrophy. Four plates/strain/dose were exposed to 0, 20, 50, 100, 200, 500, 2500 or 12500 µg/plate, in the presence or absence of metabolic activation system (S9 fraction of Aroclor 1254-induced, male Sprague-Dawley rat liver homogenates). Colonies per plate and total bacteria counts were measured. Treatment-related increased mutation frequency was not seen. No adverse effect was indicated. The study is unacceptable and not upgradeable because: repeat trials did not include all dose levels and strains, inadequate rationale for the doses and vehicle, lack of experimental details in the report, and no analytical data (J. Schreider, 1/27/85; H. Green and S. Morris, 11/24/92).

315-065; 027099: Exact duplicate of doc. # 315-061, rec. # 019920.

315-061; 019921; "Salmonella/Mammalian Microsome Mutagenicity Test (Ames Test) with Monitor Technical." (SOCAL 1711; M.L. Machado, J.A. Parker and Z.A. Wong; Chevron Environmental Health Center, Richmond, CA; 2/3/82.) Methamidophos (Monitor Technical, purity not stated, Mobay 77-297-149) was tested in a bacterial assay that measured the rate of mutation of a histidine auxotrophic strains of *Salmonella typhimurium* (tester strains TA98, TA100, TA1535, TA1537, and TA1538) to prototrophy. Three plates/strain/dose were exposed to 0, 0.1, 0.5, 1.0, 5.0, or 10 µg/plate, in the presence or absence of metabolic activation system (liver homogenate S-9 fraction, no other details). Colonies per plate were measured. Treatment-related increased mutation frequency was not seen. No adverse effect was indicated. The study is unacceptable and not upgradeable because: no repeat trials, lack of experimental details in the report, inadequate rationale for the doses, inadequate analytical data, and no cytotoxicity data (J. Schreider, 1/25/85; H. Green and S. Morris, 11/25/92)

315-018; 960216: Exact duplicate of doc. # 315-061, rec. # 019921.

315-040; 960217: Exact duplicate of doc. # 315-061, rec. # 019921.

CHROMOSOME EFFECTS

** 315-110; 090583; "Mutagenicity Test on SRA 5172 In An In Vitro Cytogenetic Assay Measuring Chromosomal Aberration frequencies in Chinese Hamster Ovary (CHO) Cells." (HLA Study No. 10972-0-437; M. Hemalatha; Hazleton Laboratories America, Inc., Kensington, MD; 1/19/90.) Methamidophos (SRA 5172, 74.5% stated purity) was tested in vitro for induction of chromosome aberrations in cultured Chinese hamster ovary (CHO) Cells. Exponentially-growing monolayers were treated in duplicate for varying times with the test material at 0, 1870, 2500, 2570, 3150, 3850, 4200, 5140, or 5250 µg/ml without or with S-9 metabolic activation system (9000 x g supernatant of Aroclor 1254 induced, male Sprague-Dawley rat liver homogenates) at 0, 1250, 3750, or 4990 µg/ml. The cells were washed 2.5 hours before harvest and re-incubated in medium with 0.1 mg/ml Colcemid. A **possible adverse effect** was indicated by treatment-related increases in chromosome aberrations with or without activation. The study is acceptable (H. Green and S. Morris, 12/30/92)

315-061; 019923 "In Vivo Cytogenetics Study in Mice, Methamidophos Technical (SX-1244)." (MRI-176-CCC-82-36; H.J. Esber, EG&G/Mason Research Institute, Worcester, MA; 11/18/83.) Methamidophos Technical (SX-1244, batch # 77-297-149, 73.5% stated purity) was given in water by single oral gavage at 0.0, 0.8, 2.7, 8.1, 12.1, or 16.1 mg/kg to 12 CD1 mice/sex/group. Mice were given colchicine ip at 1.2 mg/kg 2 hours prior to sacrifice. At 6, 24, or 48 hours after dosing, 4 mice/sex/dose were sacrificed and femoral bone marrow samples were fixed and stained and 50 metaphase cells/mouse were analyzed microscopically for chromosome aberrations. There were no treatment-related effects on chromosome aberrations. No adverse effect was indicated. A NOEL for anticholinesterase activity of < 4.1 mg/kg was based on clinical signs in a preliminary acute mortality studies. The study was unacceptable because of inadequate rationales for doses and sampling intervals and no analysis of dosing solutions and not upgradeable because there were inadequate mice/sex/dose/time point (J. Schreider, 1/30/85; H. Green and S. Morris, 12/21/92).

315-045; 020163: Exact duplicate of doc. # 315-061, rec. # 019923.

315-045; 020164: Protocol for study at doc. # 315-061, rec. # 019923.

315-061; 019919 "Dominant Lethal Study of Methamidophos Technical in Mice." (SOCAL 1783; G.H. Eisenlord, J.H. Carver and Z.A. Wong; Chevron Environmental Health Center, Inc., Richmond, CA; 3/23/84.) Methamidophos Technical (Mobay Reference No. 77-297-149, 74.3% stated purity) was fed in the diet of groups of 12 CD-1 male mice for 5 days at 0, 5, 50, or 150 ppm followed by an 8-week mating period in which each male was paired with 2 new females every week for a total of 16 females per male. Eight days after each 1-week mating period, the females were sacrificed and their uteri were examined for the numbers of implants, live fetuses and early and late fetal deaths. At the end of the 8-week mating period, the males were sacrificed and necropsied. Group mean food consumption for the 5 treatment days and body weight on day 5 were reduced in the 150 ppm males to respectively 53% and 88% of controls (subacute NOEL = 4.6 mg/kg/day). There were no other treatment-related effects reported on males or uterine variables. No adverse effect was indicated. The study is unacceptable because of an inadequate rationale for the doses used and not upgradeable because of an inadequate number of pregnant females (J. Schreider, 1/29/85; H. Green and S. Morris 12/16/92)

315-065; 027097: Partial duplicate of doc. # 315-061, rec. # 019919.

315-061; 019918; "Dominant Lethal Test on Male Mouse to Evaluate SRA 5172 for Mutagenic Potential." (Report No. 9583; Dr. B. Herbold, Bayer AG, Institut Für Toxikologie, Wuppertal, Germany; 11/26/80.) Methamidophos (SRA 5172, 62.6% stated purity) was emulsified in 0.5% Cremophor and given by a single oral gavage to groups of 50 male NMRI/ORIG Kissleg mice at 0 or 5 mg/kg on day 1. Starting on day 1 each male mouse was paired with a new female mouse every 4 days for 48 days for a total of 12 females/male mouse. The females were sacrificed 12 days after the pairing interval and their uteri were examined for total implants, viable implants, dead implants, and Corpora lutea. There were no treatment-related effects on the males body weight, appetite, physical appearance, motor activity, or survival. There were no treatment-related effects on uterine variables. No adverse effect was indicated. The study is unacceptable because the rationales for the doses and vehicle were inadequate and there was no analysis of dosing material. The study is not upgradeable because there was no positive control (J. Schreider, 1/29/85; H. Green and S. Morris, 12/11/92).

315-065; 027098: Partial duplicate of doc. # 315-061, rec. # 019918.

315-061; 019922 "Micronucleus Test on the Mouse to Evaluate for Mutagenic Effect." (Report No. 9707; B. Herbold; Bayer AG, Institute of Toxicology, Wuppertal-Elberfeld, Germany; 1/22/81) Methamidophos (SRA 5172, 62.6% stated purity) was given by oral gavage (0.5% Cremophor suspension) to five NMRI/W 77 mice/sex/group at 0, 5, or 10 mg/kg. Two doses were given 24 hours apart and femoral bone marrow samples were taken 6 hours after the second dose. For each mouse, 1,000 polychromatic erythrocytes were scored for micronuclei and the ratio of normo/poly chromatic erythrocytes was determined. Convulsions were observed in one mouse/sex at the high dose. There were no other treatment-related effects. No adverse effect was indicated. The study is unacceptable but possibly upgradeable with adequate submissions of analysis of the dosing material and rationales for the dose levels, timing, and vehicle (J. Schreider, 1/30/85; H. Green and S. Morris, 12/1/92).

315-065; 027096: Exact duplicate of doc. # 315-061, rec. # 019922.

DNA DAMAGE

**315-153;149102; "SRA 5172 Micronucleus Test on the Mouse," (Study No. T 9060076, Bayer Report No. 107443; B. Herbold; Bayer AG, Wuppertal, Germany; 5/23/96.) Groups of 5 Hsd/Win:NMRI mice / sex / time point were given single intraperitoneal injections of methamidophos (SRA 5172, batch # 278467030, 75% analytical purity, saline vehicle, 10 ml/kg) at 8 mg/kg. Femoral marrow samples were taken 16, 24, or 48 hours later. Marrow smears were dried and stained. One thousand polychromatic erythrocytes / animal were microscopically evaluated for micronuclei, and the number of normochromatic erythrocytes / 1000 polychromatic erythrocytes and the number of normochromatic erythrocytes with micronuclei were determined. There was no treatment-related effect on the micronuclei incidence. No adverse effect was indicated. The positive controls were adequate. The study was acceptable (S. Morris and J. Gee, 5/6/97).

315-153; 149102; p. 13. A brief abstract of a pilot study was included in the main report above. Groups of both sexes of 5 mice/dose were given intraperitoneal injections of methamidophos at 8,10, or 50 mg/kg. Animals at all doses exhibited apathy, roughened

fur, sternal recumbency, spasm, palmo-spasm, difficulty in breathing, eyelids stuck together, lachrymation, and salivation. One of 5 and 5 of 5 mice died at 10 and 50 mg/kg, respectively. These data are an adequate rationale for the dose used in the main study.

315-061; 019924; "Pol Test on E. Coli to Evaluate for DNA Damage." (Report No. 12318; Dr. B. Herbold, Bayer AG, Institute of Toxicology, Wuppertal-Elberfeld; 12/19/83.) Methamidophos (SRA 5172, batch 808319101, 71.2% stated purity) was tested for genotoxic activity in an assay that compared the inhibition of growth of two strains of the bacterium Escherichia coli. One strain was deficient ((K12)p 3478) while the other was proficient (W 3110) in DNA repair. The assay was conducted with or without metabolic activation (S-9 fraction of Aroclor-1254-induced, male rat liver homogenates) at 0, 625, 1250, 2500, 5000, or 10000 µg/plate. The plate diffusion method was used with 4 plates/dose/ strain being incubated for 24 hours. No adverse effect was indicated. The study is unacceptable because there was no rationale for the doses used, no individual plate data, no viability data, no concentration or rationale for the vehicle, and the report did not adequately describe the protocol. The study is not upgradeable because there was no positive control for metabolic activation and the 2 strains were not equally sensitive to the negative control agent (J. Schreider, 1/30/85; (H. Green and S. Morris 12/4/92).

315-065; 027095: Exact duplicate of doc. # 315-061 rec. # 019924.

315-105; 075732; "Unscheduled DNA Synthesis in Rat Primary Hepatocytes." (MBA Study No. T5844.380; R.D. Curren, Microbiological Associates, Inc., Rockville, MD; 10/24/88.) Methamidophos (Monitor Technical, Reference No. 77-297-149, 71.2% stated purity, DMSO solvent) was tested in an unscheduled DNA synthesis (UDS) assay. Triplicate plates with primary male Sprague-Dawley rat hepatocytes attached to coverslips were incubated for 18-20 hours in medium containing the test material at 0, 0.001, 0.003, 0.01, 0.03, 0.1, 0.3, 1.0, 3.0, or 10.0 µl/ml and 3H-thymidine. The cells were fixed, developed for autoradiography and stained for cellular material. Nuclear grain counts were made for 50 cells/plate. There was no treatment-related effect on net nuclear grain counts. No adverse effect was indicated. The study is unacceptable but possibly upgradeable with submissions of adequate cytotoxicity data, protocol for the LDH assay, rationales for the doses and solvent, raw grain counts, specific activity of 3H-thymidine, and the morphological criteria and rationale for excluding cells from nuclear grain counts (H. Green and S. Morris 1/5/93).

NEUROTOXICITY, HENS

**315-146; 141419; "Subchronic Dermal Neurotoxicity Study (Ninety-Day Hen Study)," (Study No. T1033771, Bayer No. 105085; W. Bomann, G. Kaliner and H. Mager; Bayer AG, Wuppertal, Germany; 6/25/93.) Groups of 15 to 25 leghorn hens were dermally exposed to methamidophos (SRA 5172, analytical No. 31192, Fs 23280, 76.3% analytical purity, isopropanol solvent, 0.02 ml/kg b.w.), once a day, 5 days per week for 13 weeks at 0, 0.5, 1.5, or 4.5 mg/kg/day. Neuropathy target esterase (NTE) level was determined in brain and spinal cord homogenates from 1 to 3 hens per group, 24 hours after the last treatments of weeks 4 and 13. Clinical appearance and behavior were monitored once daily and forced activity and ladder climbing tests were performed weekly for all groups during treatment and 4 weeks post treatment for the 0 and 4.5 mg/kg/day groups. Plasma cholinesterase levels (ChE) were measured on 10 birds/group 1 week prior to treatment and 24 hours after the last treatments of weeks 3, 6, and 13 and week 17 (4 weeks post treatment, 0 and 4.5 mg/kg/day only). All birds

were sacrificed at the end of their treatment or post-treatment observation period. Sciatic nerve, brain and spinal cord were examine histologically. TOCP was the positive control. Treatment-related effects seen at 4.5 mg/kg/day were apathy, ruffled feathers, staggering gait, reduced feed intake, transient decrease in body weight, discolored feces, and diarrhea. Detachment of the uppermost layer of skin at the treatment site was seen at 1.5 and 4.5 mg/kg/day. One animal in each treatment group either died or was sacrificed moribund. There were no treatment-related effects on forced activity or ladder climbing. At 4 and 13 weeks, brain and spinal cord NTE were decreased at 4.5 mg/kg/day. At weeks 3, 6, and 13, plasma ChE was decreased at 1.5 and 4.5 mg/kg/day. There were no treatment-related histopathology findings. The positive control was adequate. There was no treatment-related neuropathy. No adverse effect was indicated. The study was acceptable (S. Morris and J. Gee, 4/7/97).

315-013; 960205; "Neurotoxicity Study - Chickens Monitor RE 9006, 75 Percent Technical"; IBT No. J6480; 11/12/68. Invalid IBT study (J. Schreider, 1/28/85).

315-115; 095393; Acute Delayed Neurotoxicity; 817; Hen; Bayer AG, Dept. of Toxicology, Wuppertal, Germany; Mobay Report No. 100281; 4/10/90; 3 test articles: #1. (+) Methamidophos, #2. (+) Methamidophos, #3. (-) Methamidophos; Study No. T3029958: #2, (100 and 200 mg/kg)-5 hens/dose; T2029722: Control-11 hens; #2, #3 (400 mg/kg)-13 hens; TOCP (100 mg/kg)-2 hens; T2029957: #3 (400 mg/kg)-10 hens; T1029956: #1 (200 and 400 mg/kg)-10 hens/dose; Mortalities: T3029958: #2-0/10; T2029722: Control-0/11, #2-3/13, #3-6/13, TOCP-0/2; T2029957: #3-9/10; T1029956: #1-2/10 (200 mg/kg), 7/10 (400 mg/kg), all deaths within 6 days post-dosing, except for 1 hen which died day 28 (T1029956, 200 mg/kg); Clinical signs: acute phase (common to three test compounds, all dose groups)-apathy, ruffled feathers, staggering gait, diarrhea, rapid shallow breathing, some cases of flat, lateral prostration, spasms, (in addition for #3) salivation, labored breathing, dry and limp comb; OPIDP: T3029958-100 mg/kg, no signs, 200 mg/kg-abnormal gait, reversible; T2029722-#2 (400 mg/kg) 2 totally paralyzed by day 18, 1 marked ataxia, 1 ataxic, disturbed motor coordination; #3 (400 mg/kg) no signs (1 hen); TOCP-treated not observed; T2029957-#3 (400 mg/kg) no signs (1 hen); T1029956-#1 (200 mg/kg) no signs (9 hens), (400 mg/kg) 1 disturbed motor coordination, 2 slightly abnormal gait; Necropsy: (animals which survived the acute phase) pale, sometimes lobulated liver ((+)-Methamidophos, 200 and 400 mg/kg, 7/11 hens); No histopathology performed. Unacceptable and not upgradeable (No histopathological evaluation of the target tissues was performed.) (T. Moore, 11/28/90).

315-027; 000046. This document contains a brief summary of a study in which an unspecified number of full-grown hens were injected ip with PAM (0.1 g/kg) and atropine sulphate (0.05 g/kg) followed by oral (50 or 100 mg/kg) or ip (25, 50 or 100 mg/kg) exposure to the active ingredient and observed for 42 days. No neurotoxic effects were reported. No worksheet was done (S. Morris, 9/16/92).

315-027; 001212 "Acute Delayed Neurotoxicity Study on Monitor Technical." (Study No. ANHO1, Mobay No. 68037, S.M. Kruckenberg et al., Department of Pathology, College of Veterinary Medicine, Kansas State University, Manhattan, KS, 7/29/79.) Adult White Leghorn hens were simultaneously given im injections of atropine sulfate at 50 mg/kg and Methamidophos (Monitor Technical, 74% stated purity, batch no. 9030005, analysis not stated, water vehicle) by oral gavage at 0.00 (8 hens), 30.00 (10 hens), or 50.63 (12 hens) mg/kg on days 0 and 21 and observed for 42 days. There were 2/10 and 4/12 lethality at 30.00 and 50.63 mg/kg respectively. There were no clinical signs or histological evidence in spinal cord and sciatic nerve of delayed neuropathy. The rationale for dosing was adequately based on a

preliminary lethality study. The positive controls were adequate. No adverse effect was indicated. The study was unacceptable but possibly upgradeable by submission of adequate analysis of the test material (J. Schreider, 1/29/85; H. Green and S. Morris, 9/18/92).

315-029; 001215: Partial duplicate of doc. # 315-027, rec. # 001212.

315-050; 017046: Exact duplicate of doc. # 315-027, rec. # 001212.

315-067; 028429: Exact duplicate of doc. # 315-027, rec. # 001212.

315-001; 960201: Partial duplicate of doc. # 315-027, rec. # 001212.

315-031; 960202: Partial duplicate of doc. # 315-027, rec. # 001212.

315-061; 019925; "Methamidophos (Tameron Active Ingredient) and Tameron, Sri Lanka Formulation, Special Study for Neurotoxic Effects on the Chicken." (J. Thyssen and A. Eben, Bayer AG Institute of Toxicology, Report No. 10815, 4/20/82.) Two formulations of Methamidophos (Tameron technical BR, batch 808018244, 74.6% stated purity and Tameron Fl. 1376/476, Sri Lanka formulation, 52.4% stated purity) were tested. There were 4 protocols that differed in: test material, dose, dosing schedule, numbers of hens, post-treatment clinical observation period, serial sacrifice schedule, and testing for neurotoxic esterase activity (NTE). Groups of 5 to 30 adult white Leghorn hens were treated on 5 consecutive days by oral gavage with test material at 25, 30, or 35 mg/kg (water vehicle, doses corrected for purity) and simultaneous im injections of atropine sulphate at 50 mg/kg. All protocols produced acute mortality and all hens displayed clinical cholinergic signs for up to 15 days post-treatment. Brain, spinal cord and sciatic nerve levels of NTE were severely depressed on post-treatment day 1 but gradually recovered on days 2, 3 and 5 to normal levels on day 38. Delayed neurotoxicity was not observed at 42 days post-treatment. No adverse effect was indicated. The study was not a standard neurotoxicity protocol, was therefore considered supplemental information and no updated worksheet was done (J. Schreider, 1/30/85; S. Morris and H. Green, 10/6/92).

315-064; 027091: Exact duplicate of doc. # 315-061, rec. # 019925.

315-067; 028431: Exact duplicate of doc. # 315-061, rec. # 019925.

315-013; 960205; "Neurotoxicity Study - Chickens Monitor RE 9006, 75 Percent Technical", IBT No. J6480; Invalid IBT study; no worksheet (J. Schreider, 1/28/85; S. Morris, 10/9/92).

315-115; 095394; NTE Assay after Oral Administration; Hen; Bayer AG, Dept. of Toxicology, Wuppertal, Germany; Mobay Report No. 100280; 5/29/90; 3 test articles: #1. (±)-Methamidophos, #2. (+)-Methamidophos, #3. (-)-Methamidophos; Study No. T3027743: Control (6 hens), #1 (50 mg/kg) (6), TOCP (300 mg/kg) (4), NTE assayed 24, 48 hrs post-dosing, lymphocyte, brain, spinal cord, sciatic nerve; T3029543: Control (6), #2 (50 mg/kg) (6), #3 (50 mg/kg) (6), TOCP (100 mg/kg) (2), NTE assayed 24, 48 hours post-dosing, brain; T2029722: Control (6), #2 (400 mg/kg) (6), #3 (400 mg/kg) (6), TOCP (300 mg/kg) (2), NTE assayed 24, 48 hrs post-dosing, brain; T4030335: Control (3), #2 (400 mg/kg) (6), NTE assayed 24 hrs, brain; T6032001: Control (8), #2 (100 mg/kg) (9), #3 (200 mg/kg) (9), NTE assayed 24, 48 hrs and 7 days, lymphocyte, brain, spinal cord, and sciatic nerve; Results: #2 and #3-dose-dependent % inhibition of brain NTE; % inhibition (50 mg/kg)-#1=#2>#3; spinal cord, sciatic nerve % inhibition equal to brain; lymphocyte-activity more quickly recovered; reactivation of NTE-#1=#2>#3;

TOCP (positive control)-90 to 100% inhibition, minimal reactivation. Supplemental (T. Moore, 11/29/90)

315-102; 070877; "3-Month Subchronic Delayed Neurotoxicity Study with SRA 5172 (C.N. Methamidophos)" K. Sachse et al.; KFM Kleintierfarm Madoerin AG, Fuellinsdorf, Switzerland; RCC Research and Consulting Company AG, Itingen, Switzerland; and RCC Umweltchemie AG, Itingen, Switzerland; Laboratory Project ID 94213/064293; 5/15/87. Methamidophos (SRA 5172, batch 808 526 298, 76% stated purity) was given 5 days/week for 3 months by oral gavage (water vehicle) at 0, 0.3, 1.0, or 3.0 mg/kg/day to groups of 16 White Leghorn hens per dose. Motor activity of each hen was measured twice weekly. Plasma cholinesterase levels were measured after 4, 8, and 12 weeks of treatment and central and peripheral neuro-histopathology done at termination on 10 hens/ dose. Terminal brain and spinal cord neurotoxic esterase (NTE) was measured on the remaining 6 hens/dose. Mortalities, 2 each at 0 and 3.0 mg/kg, were not treatment-related. Treatment-related effects were: somnolence and terminal group mean body weight was 80% controls at 3.0 mg/kg, 12-week plasma cholinesterase levels were 83 and 56% of controls respectively at 1.0 and 3.0 mg/kg, brain NTE was 83% of controls at 3.0 mg/kg, and spinal cord NTE was 78 and 59% of controls respectively at 1.0 and 3.0 mg/kg. There were no behavioral or histopathological indications of delayed neurotoxicity. No adverse effect was indicated. This study was not a required test type and was therefore not evaluated for acceptability and no worksheet was done (H. Green and S. Morris, 10/8/92).

SUPPLEMENTAL (RATS, HUMANS)

**315-122; 089116; "SRA 5172 Study of the Subchronic Inhalation Toxicity to Rats in Accordance with OECD Guideline No. 413." (Laboratory Project ID Report No. 98370; J. Pauluhn, Bayer AG, Wuppertal, Germany; 3/30/88.) Groups of 10 Wistar rats/sex were head/nose exposed 6 hours/day, 5 days/week for 3 months to methamidophos (SRA 5172, batch # TOX 1767-00, 73.4%) at mean analytical concentrations of 0 (air only), 0 (vehicle), 0 (vehicle, recovery group), 1.1, 5.4, 23.1 (recovery group), or 23.1 mg/m³. After the 3 month exposure period, the two recovery groups (vehicle and 23.1 mg/m³) were allowed a 6-week exposure-free period. Treatment-related effects were seen in both sexes at 23.1 mg/m³: slight to moderate tremors on the day of exposure but not prior to exposure the next day; decreased body weight gain, decreased relative spleen weights and increased relative adrenal weights. There were treatment-related, noncumulative decreases in cholinesterase activity in plasma and brain, and increased sensitivity to the acetylcholine provocation test in both sexes at 5.4 and 23.1 mg/m³. The NOEL = 1.1 mg/m³ (0.3 mg/kg body weight/day) based on decreases in brain cholinesterase at the mid and high doses and tremors at the high dose. No adverse effect was indicated. The study is acceptable as supplemental data (S. Morris and J. Gee, 1/19/99).

315-120; 089114: Supplemental data for doc. # 315-122, rec. # 089116.

315-125; 89440; "Technical Grade Methamidophos (Monitor): An Eight-Week Subchronic Cholinesterase Study in Fischer 344 Rats" 855; Rat; Mobay Corporation, Health, Environment, Safety and Plant Management, Corporate Toxicology Department, Stilwell, KS; Project# 100667; 3/19/91; Methamidophos; Batch No. 0067009; Doses: (nominal-0, 0.5, 1, 2, 4 ppm), 0 (vehicle-corn oil (1% w/w of diet), 0.49, 0.97, 2.12, 4.30 ppm; (M): 0.028, 0.055, 0.122, 0.244 mg/kg/day, respectively; (F): 0.033, 0.065, 0.143, 0.284 mg/kg/day, respectively; 25 animals/sex/group; No mortality; Observations: no treatment-related signs; Cholinesterase Assays: Dose-related

inhibition of plasma butyrylcholinesterase (PBChE), red blood cell acetylcholinesterase (RChE), and brain acetylcholinesterase (BChE), > 30% inhibition (4.30 ppm) F: PBChE-days 14, 42, BChE-day 56; M: PBChE-day 42; NOEL can not be determined; study unacceptable, but may be upgradeable with the submission of a more detailed analysis of the test article. (Moore, 6/18/91).

** 315-131 127242 Hamilton, B. "An Acute Oral Neurotoxicity Screening Study with Technical Grade Methamidophos (Monitor®) in Rats", (Miles, Inc., Agriculture Division, Toxicology, Stilwell, KS. Miles, Inc. Report # 105053, 11/5/93). Methamidophos technical (purity 75.6%, Batch 0-06-7009) was administered by single oral gavage to 24 Sprague-Dawley (Sas:CD(SD)BR) rats/sex/group at 0, 0.9, 3.3 and 9.0 mg/kg. Daily clinical observations (eg. muscle fasciculations, ataxia, urine stain, and nasal stain) and FOB observations (eg. gait incoordination, muscle fasciculations, salivation, urine stains, tremors, reduced rearing and reduced reflex reactions) were reflective of acute cholinesterase inhibition. Motor and locomotor activity was decreased in all dose groups at day 0; only high-dose males showed reduction in motor activity by day 7. **NOEL (clinical signs, FOB and motor activity test) < 0.9 mg/kg.** Serum aspartate amino-transferase (AST), serum alanine aminotransferase (ALT) and cholesterol values were increased in high-dose males and females. Plasma, RBC and brain cholinesterase (ChE) activity was significantly depressed at all dosage levels (up to 92% inhibition in the high-dose group) two hours after treatment. **NOEL (ChE inhibition) < 0.9 mg/kg.** No histopathological lesions; **No Adverse Neurotoxic Effects.** Originally UNACCEPTABLE; upgradeable with submission of analytical data for the test compound and positive (historical) control data (Green, Kellner and Gee, 5/6/94). This study was upgraded to **ACCEPTABLE** after review of historical positive control data in 374-087:122985. Kellner, 6/5/95.

374-087:122985: Historical positive control data used as a supplement to 315-131:127242; Sheets L.P., "Historical control and method validation studies in rats for the acute and subchronic neurotoxicity screening battery", Miles Inc., Agricultural Division, Toxicology, Stilwell, Kansas, Miles Report No. 103979, 3/31/93. This volume contains verification of the test procedures used for motor activity, FOB and neuropathology using positive control substances with known neurobehavioral and neuropathological effects. Animals were treated with chlorpromazine and triadimefon for the motor activity tests, with acrylamide and carbaryl for the Functional Observational Battery (FOB) and with acrylamide or trimethyltin for neuropathology. These data allow an upgrade of study 315-131:127242 to **ACCEPTABLE**. Another review of these data is contained in a worksheet by C. Aldous, appearing under 374-087:122985. Kellner, 6/5/95.

** 315-139 132008 Sheets, L. P. "An Acute Oral Neurotoxicity Screening Study with Technical Grade Methamidophos (Monitor®) in Rats", (Miles, Inc., Agriculture Division, Toxicology, Stilwell, KS. Miles, Inc. Report # 105053-1, 8/12/94). Methamidophos technical (purity 75.6%, Batch 0-06-7009) was administered by single oral gavage to 18 Sprague-Dawley (Sas:CD(SD)BR) rats/sex/group at 0, 0.3 and 0.6 mg/kg. There were no treatment-related effects on motor activity, body weights or daily clinical signs. Possible treatment-related FOB observations included increased landing footsplay in high-dose males. **NOEL (for neurobehavioral effects) = 0.3 mg/kg.** Gross pathological or micropathology examinations were not performed. Significant reductions in RBC, plasma and brain cholinesterase (ChE) activity in males and RBC and brain ChE in females were noted at the high-dose level. **NOEL**

(ChE inhibition) = 0.3 mg/kg. No Adverse Neurotoxic Effects. ACCEPTABLE. Kellner and Gee, 6/5/95.

315-148 142828. This document contains data supplemental to the study at DPR doc. # 315-139, rec # 132008. This document contained supplemental information on the stability, homogeneity, and purity of the test material and dosing solutions and method for measuring ChE activity. No worksheet was done (S. Morris, 2/6/96).

315-135 129816 Sheets, L. "A Subchronic Dietary Neurotoxicity Screening Study with Technical Grade Methamidophos (Monitor®) in Fischer 344 Rats", (Miles, Inc., Agriculture Division, Toxicology, Stilwell, KS. Miles, Inc. Report # 106351, 4/13/94). Methamidophos technical (purity 75.6%, Batch 0-06-7009) was administered in the diet for 13 weeks to 18 Fischer 344 rats/sex/group at nominal concentrations of 0, 1.0, 12 and 60 ppm (mean intake males: 0.067, 0.787 and 4.26 mg/kg/day; females: 0.074, 0.899 and 4.94 mg/kg/day). Daily clinical observations (eg. muscle fasciculations, increased reactivity, perianal stain, urine stain and lacrimation), FOB observations (eg. muscle fasciculations, salivation, urine stains, tremors, decreased forelimb grip strength) and decreased motor and locomotor activity were noted in the mid- and high-dose rats. High-dose rats showed reduced motor activity during weeks 4, 8 and 13 (reductions in the mid-dose females during week 4 only). **NOEL (clinical signs, FOB and motor activity test) = 1.0 ppm. Plasma, RBC and brain cholinesterase (ChE) activity was significantly depressed in the mid- and high-dose groups (up to 97% RBC ChE inhibition by week 13 in the high-dose males) **NOEL (ChE inhibition) = 1.0 ppm.** No histopathological lesions; **No Adverse Neurotoxic Effects.** Unacceptable (Kellner and Gee, 6/5/94) but upgraded to acceptable with clarification of active ingredient content of test feed (i.e. if correction made for 75.6% purity of technical methamidophos) and submission of positive (historical) control data (S. Morris and J. Gee, 5/8/97)

315-152; 149098: This document contained clarification of the active ingredient content of test feed and positive (historical) control data. Evaluation of these data resulted in upgrading the study status to adequate.

315-067; 028430. This document contains a brief review of some acute human poisonings with the test material and related animal data. A worksheet was not done (S. Morris, 10/7/92).

315-061; 028430: Exact duplicate of doc. # 315-067, rec # 028430.

315-061; 020053; N. Senanayake and M.K. Johnson (1982), "Acute Polyneuropathy after Poisoning by a New Organophosphate Insecticide", *The New England Journal of Medicine* 306:155-157, 1/27/82. This article discusses 10 human exposures (7 suicide attempts and 3 accidental poisonings) to toxic levels of methamidophos in Sri Lanka. Acute symptoms of toxicity described include unconsciousness, pupillary constriction, muscular fasciculations, and profuse sweating. A **possible adverse effect** was reported: delayed neuropathy (muscle weakness). This study was not a required test type and was therefore not evaluated for acceptability and no worksheet was done (H. Green and S. Morris, 1/6/93).

315-115; 095397; "Can Methamidophos Cause Delayed Polyneuropathy in Man or in Test Animals?"; Literature review; M.K. Johnson and M. Lotti; The authors assessed the potential of methamidophos to induce OPIDP in humans. Several instances have been reported of accidental overdoses to methamidophos in Nicaragua and Sri Lanka. OPIDP-like symptoms were observed in these patients 2 to 3 weeks after an episode of severe cholinergic poisoning.

They required a period of 6 weeks to 2 years to recover from the neuropathy. Researchers have been able to reproduce in hens a similar response with a single dose of the racemate (400 mg/kg) or a multiple dose regimen of 130 + 50 + 50 mg/kg given over a 4 day period. These doses are well in excess of the LD50 value of 25 mg/kg for the racemate and the hens required vigorous antidotal therapy. Inhibition of the target tissues in these hens was > 86%. The authors compared the *in vitro* I50 AChE/I50 NTE ratio for hens to the LD50/OPIDP value. The former ratio had a range of 0.022 to 0.064 in comparison to 0.083 for the latter value. These values indicate that more than a 10 fold concentration of methamidophos is required to inhibit 50% of the NTE activity than that of AChE and that a greater than 10 fold dose is required to induce OPIDP than to achieve the LD50. Treatment with the D-(+) isomer (200 mg/kg) was sufficient to produce signs of OPIDP which were reversible. In contrast treatment with the L-(-) isomer (400 mg/kg) was insufficient to produce any signs of OPIDP in the surviving birds (2/17).

Reactivation studies revealed that > 80% of the NTE activity inhibited with the racemate and the D-(+) isomer could be reactivated by KF. These results indicated that the enzyme had not aged, a step considered necessary for the induction of OPIDP. NTE inhibited by the L-(-) isomer could not be reactivated. The I50 AChE/I50 NTE ratio (racemate) for human brain tissue is 0.064. This value taken in conjunction with clinical observations obtained from patients suffering from an overdose of methamidophos confirms that humans are only susceptible to the induction of OPIDP at a dose quite in excess of the LD50 value. (T. Moore, 4/26/91).

315-126; 089467: Exact duplicate of doc. # 315-115, rec. # 095397.

315-102; 070878; "The Cholinesterase Inhibition Potential of Analytical Grade Methamidophos (SX-1672) and Methamidophos Technical (SX-1490) Following Topical Application of a Single Dose to Male and Female Rats." Study No, S-2284; M.D. Easter and D.W. Rosenberg; Chevron Environmental Health Center, Inc., Richmond, CA; 6/27/86. Groups of 5 Sprague-Dawley rats/sex were given analytical (SX-1672, 99.1% purity) or technical (SX-1490, 74.7% purity) grade Methamidophos by single dermal applications to shaved dorsal skin at 0, 1.00, 2.50, 6.25, and 15.60 mg/rat. Animals were sacrificed 24 or 72 hours later and brain, red blood cell (RBC), and plasma cholinesterase levels (ChE) were measured. With the analytical grade, RBC, plasma, and brain ChE were inhibited at 1.00 mg/rat in females at 24 hours and in both sexes at 2.50 mg/rat after 72 hours. With the technical grade RBC, plasma, or brain ChE were inhibited in both sexes at 2.50 mg/rat at 24 and 72 hours. No adverse effect was indicated. This study was not a required test type and was therefore not evaluated for acceptability and no worksheet was done (H. Green and S. Morris, 1/5/93).

315-158; 160096 "Repeated-Dose 21-Day Dermal Toxicity Study with Technical Grade Methamidophos (Monitor®) in Rats," (L.P. Sheets and M.E. Gastner; Bayer Corporation, Stilwell, KA; Study Number 96-122-KQ, Report Number 107635, 12/10/97.) Methamidophos (batch # 703-0001, 76.9 to 80.5% purity) was applied as aqueous solutions (1 ml/kg) daily for 6 hours to the shorn backs of groups of 9 or 10 Sprague-Dawley rats/sex/dose for 18 of 22 days (males) or 17 of 21 days (females) at nominal doses of 0, 1, 15, or 50 mg/kg/day (analytical doses were 0, 1, 15, 47 mg/kg/day). Observations and measurements were made: detailed clinical observations, body weight, food consumption, ophthalmology, brain, erythrocyte (RBC) and plasma cholinesterase (ChE) activity, organ weights, gross necropsy findings and micropathology. Brain, RBC, and plasma ChE activities were decreased at 15 at 50 mg/kg/day (ChE inhibition NOEL = 1 mg/kg/day). Males had the greatest depression of brain ChE which was 34% of controls. There were no other treatment-related effects reported. No adverse effect was indicated. The study is unacceptable but possibly upgradeable with adequate submissions of analytical data for the test material and dosing solutions; hematology, serum chemistry, and

an adequate rationale for the highest dose (S. Morris and J Gee, 4/15/98). See record 206823 in 315-169 for adequate analytical data for record 160096, satisfying that deficiency. The study remains unacceptable based on the lack of hematology and clinical chemistry. (Gee, 10/1/03)

315-169 206823 Supplemental to 160096. Moore, K. D., 9/28/98, Report Number 107635-1. This supplement presents the analytical data for the dermal study. The active ingredient content of the dose preparations was actually 0.749, 11.2 and 36.5 mg of methamidophos per ml. The dose preparations were stable in the refrigerator for 21 days. In addition, five batches of technical material were analyzed and 30 compounds identified in each batch. The average content of methamidophos was 76 - 77% of the 98% total identified. No worksheet. (Gee, 10/1/03)

315-169 206824 Duplicate of 160096 in 315-158.

315 - 0167 200767 " A developmental neurotoxicity screening study with technical grade methamidophos (Monitor) in Wistar rats." (Sheets, L. P., Pathology Report by S. G. Lake, Bayer Corp., Agriculture Division, KS, Study No. 00-D72-A1, Report No. 110924, February 11, 2002). Female Wistar CrI:W(HAN)BR rats, 30/group, were mated with untreated males, 1:1. Beginning on GD 0, females were exposed to feed containing methamidophos (batch 803-0182, 72.3 to 74.2% active ingredient) at 0, 1.0, 10 or 30 ppm (analytical concentrations were 0, 0.851, 9.77 and 27.2 ppm, adjusted for purity). Exposure continued until lactation day 21 with selected pups continuing on control diet until approximately 75 days of age. Average consumption of test article for dams during gestation: 0, 0.1, 0.9 and 2.5 mg/kg/day; during lactation: 0, 0.2, 2.4 and 7.9 mg/kg/day. Dams: There were no effects on reproduction parameters, no deaths, no treatment-related findings in the observational battery for dams on GD 6, GD 20, LD 11 and LD 21. For offspring, preputial separation was delayed at 30 ppm, body weights were lower at 30 ppm for both sexes and at 10 ppm for females. Motor activity was reduced at 10 and 30 ppm on PND 13 relative to controls but not at later times (days 17, 21 and 60). There were no treatment-related findings for acoustic startle habituation, water maze, ophthalmology, brain weight or brain morphometry or micropathology of brain or neural tissues.

Brain weights were not affected. The most significant findings concerned inhibition of cholinesterase. Plasma, RBC and brain cholinesterase inhibition were determined on lactation day 21 for the dams, PND 4 for pups (males and females combined) and PND 21, sexes separate. There was a significant inhibition of brain ChE (8%) in dams at 1.0 ppm, and in all three cholinesterase activities at 10 ppm and 30 ppm, with slightly greater inhibition at 30 ppm. For pups, day 4, at 10 ppm, RBC cholinesterase was inhibited 20% compared with controls and at 30 ppm, all three activities were significantly lower than controls. At postnatal day 21, there was no significant inhibition at 1.0 ppm for any measurement. At 10 ppm, for male pups, both plasma (-22%) and brain (-13%) ChE were significantly lower and for female pups, only brain was inhibited (-17%). At 30 ppm, all three cholinesterase activities were significantly lower in both sexes, with inhibition being slightly greater in female pups. The percent inhibition, however, for the pups was considerably less than for the dams. NOEL for dams was < 1.0 ppm (inhibition of brain ChE). NOEL for pups = 1.0 ppm (cholinesterase inhibition, lower body weight in females at 10 ppm - the only measured effect that persisted to termination). Unacceptable but upgradeable (identification and/or submission of the cited positive control studies.) No evidence of neuropathology. (Gee, 9/27/02)

315 - 0281 211501 "A study of the effects of Orthene and Monitor on plasma and erythrocyte cholinesterase activity in human subjects during subacute oral administration." (Garofalo, M., Industrial Bio-Test Laboratories, Inc., IBT No. 636-02498, Report No. 98473, March 7, 1973) Note: This study has been evaluated by US EPA as "S" for supplementary and not as "I", invalid. Pages 49 and following contain evaluations of the study made in 1977/1978, comparing the report with the available raw data. The major problem was the lack of some raw data to support the values in the IBT report, especially for the 0.4 mg/kg/day females.

Study: The test materials were mixtures of methamidophos (Monitor) and acephate (Orthene) in ratios of either 1:9 or 1:4 parts of Monitor/Orthene. The materials were taken three times daily in corn oil in gelatin capsules for daily doses of 0.1, 0.2, 0.3 or 0.4 (females only) mg/kg/day. The subjects were seven male and seven female volunteers with 2/sex in the control and 1:4 groups and 3/sex in the 1:9 group. Ages ranged from 21 to 48 years. Exposure was for a total of 21 consecutive days for 0.1, 0.2 and 0.3 mg/kg/day and 10 (?) days for 0.4 mg/kg/day in females. Baseline plasma and erythrocyte cholinesterase activities were determined 5 times during the 2 weeks preceding exposure. ChE activities were determined on days 1, 3, 7, 14 and 21 during the test period. Each subject was given increasing doses of the test materials, same ratio, in sequence of increasing dose. After exposure to 0.3 mg/kg/day, there was a 7-day rest period with evaluation of ChE activities. ChE was determined by an AutoAnalyzer using the procedure of Levine, J. B. *et. al.* Limited hematology parameters were also evaluated pretest and at the end of the exposure period. Additional observations included blood pressure, muscle tone, pulse rate, pupil size, light reflex, eye accommodation, knee jerk, tongue tremor and finger tremor. Subjects were also to report any abnormal symptoms. Results There was no effect on erythrocyte ChE in any group. There was no effect on ChE at 0.1 mg/kg/day with either ratio. At 1:4, 0.2 mg/kg, plasma ChE was depressed in both sexes (considered to be the minimum effect level by the author) but not at 1:9 ratio. At 1:9, 0.3 mg/kg/day caused depression in plasma ChE in males [1:4 was not tested at this dose]. At 0.4 mg/kg/day, 1:9, in three females, plasma, but not erythrocyte, ChE was depressed. ChE was considered affected if there were two consecutive measurements with depression greater than 2 standard deviations below the mean pretest value. The report states that there were no significant effects on hematology or the other parameters evaluated. Individual data were presented for hematology and clinical chemistry. Corrected pages, based on raw data, are included in the reevaluation pages. Supplemental study. No worksheet. (Gee, 6/18/04)