UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

014649

OFFICE OF
PREVENTION PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

Date: August 2, 2001

Subject: **Mesotrione** Registration Toxicology Disciplinary Chapter.

PC Code: 122990
DP Barcodes: D259369, D268072
Submission Nos.: S541375, S583634

From: David Nixon, Toxicologist David Nixon 8/2/2001
Registration Action Branch 1
Health Effects Division (HED) (7509C)

To: James Stone, Team 23
Joanne Miller, PM
Registration Division (7505C)

Through: George Herndon, Branch Senior Scientist A. Jeffrey Herndon
Registration Action Branch 1
Health Effects Division (HED) (7509C)

CONCLUSIONS

In the "Toxicology Disciplinary Chapter for the Registration Support Document," the Health Effects Division (HED) has evaluated the toxicity database of mesotrione and provided the Data Evaluation Records (DERs). See *Section 10 Attachments* for the list of all attached new DERs. This document also summarizes the selected toxicity endpoints and recommendations that were made by the Hazard Identification Assessment Review Committee (HIARC) and the Food Quality Protection Act (FQPA) Safety Factor (SF) Committee.

The existing toxicity database for mesotrione supports a full Section 3 registration and the establishment of permanent tolerances for residues of mesotrione in/on field corn.

The HIARC concluded that:

1) there is evidence of increased susceptibility of rats, mice and rabbits to *in utero* exposure to mesotrione and increased susceptibility of rats and mice to post-natal exposure to mesotrione;
2) a developmental neurotoxicity study (DNT) is required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects.

The FQPA Safety Factor Committee recommended that the FQPA Safety Factor be retained (10x) in assessing the risk posed by mesotrione.

ACTION REQUESTED

Syngenta Crop Protection Inc. (formerly Zeneca Ag. Products) submitted a petition for a Section 3 registration of a 4 lb/gal suspension concentrate (SC) flowable formulation (Product name = CallistoTM Herbicide) and establishment of permanent tolerances for residues of the herbicide mesotrione [2-[4-(methylsulfonyl)-2-nitrobenzoyl]-1,3-cyclohexanedione] (designated by the company code ZA1296) *per se* in/on the following raw agricultural commodities (RACs):

Field corn	0.01 ppm
Field corn, fodder	0.01 ppm
Field corn, forage	0.01 ppm

In support of these requests, a battery of acute toxicity studies on the technical grade product and on the formulated product and an extensive series of additional subchronic, chronic, developmental, reproductive, mutagenic and other toxicity studies on the technical grade product were submitted.

HED has been requested to review the toxicity database for mesotrione to determine whether it is adequate to support the registration of mesotrione in/on field corn.

BACKGROUND

Mesotrione is a triketone herbicide that inhibits the enzyme p-hydroxyphenylpyruvate dioxygenase (HPPD), disrupting carotenoid biosynthesis. This process leads to the destruction of chlorophyll, resulting in a bleaching effect in susceptible plants. Mesotrione is intended for preemergence and postemergence use for selective control of annual broadleaf weeds. There are no existing tolerances, uses, or exemptions for mesotrione. The field corn petition represents the first proposed use for mesotrione. There are currently no registered or proposed residential uses of mesotrione.

On March 13, 2001, the HIARC reviewed the recommendations of the toxicology reviewer for mesotrione with regard to the acute and chronic Reference Doses (RfDs) and the toxicological endpoint selection for occupational/residential exposure risk assessments. The potential for

increased susceptibility of infants and children from exposure to mesotrione was also evaluated as required by the Food Quality Protection Act (FQPA) of 1996. The FQPA Safety Factor Committee met on April 16, 2001 to evaluate the hazard and exposure data for mesotrione and recommended that the FQPA Safety Factor (as required by Food Quality Protection Act of August 3, 1996) be retained (10x) in assessing the risk posed by this chemical. Reports of the HIARC meeting and of the FQPA Safety Factor Committee meeting have been presented in separate memoranda [1,2]. Included in this memorandum are the Toxicology Disciplinary Chapter for the Registration Support Document and DERs for all the submitted toxicology studies (to date) on mesotrione.

cc without attachments: D. Nixon (RAB1), G. Herndon (RAB1)

---

[1]Memo, D. Nixon, Mesotrione - Report of the Hazard Identification Assessment Review Committee, 4/12/01; HED Doc. No. 014536.

[2]Memo, B. Tarplee, Mesotrione - Report of the FQPA Safety Factor Committee, 4/30/01; HED Doc. No. 014552.

MESOTRIONE (ZA1296)

PC Code: 122990

Toxicology Disciplinary Chapter for the Registration Support Document

Date completed: July, 2001

Contract Number: 68W99014
Work Assignment: 3-02-139

Prepared for:
Health Effects Division
Office of Pesticide Programs
U.S. Environmental Protection Agency
Arlington, VA 22202

Prepared by:
Pesticide Health Effects Group
Sciences Division
Dynamac Corporation
2275 Research Boulevard
Rockville, MD 20850-3268

Primary Reviewer:
Mary L. Menetrez, Ph.D.
Signature: Mary L Menetrez
Date: 7/3/01

Secondary Reviewer:
Ronnie J Bever, Ph.D.
Signature: Mary L Menetrez for Ronnie J Bever
Date: 7/3/01

Project Manager:
Mary L. Menetrez, Ph.D.
Signature: Mary L Menetrez
Date: 7/3/01

Quality Assurance:
Steve Brecher, Ph.D.
Signature: Steve Brecher
Date: 7/3/01

Disclaimer

This data summary may have been altered by the Health Effects Division subsequent to signing by Dynamac Corporation personnel.

EPA Reviewer: David Nixon
Registration Action Branch 1/HED (7509C)

EPA Work Assignment Manager: Sanyvette William-Foy
Registration Action Branch 2/HED (7509C)

David Nixon 8/2/2001

Sanyvette Foy 8/2/2K1

TABLE OF CONTENTS

1.0 HAZARD CHARACTERIZATION

Mesotrione is a triketone herbicide with a primary mode of action that inhibits the enzyme 4-hydroxyphenylpyruvate dioxygenase (HPPD), an enzyme that is integral in the catabolism of tyrosine in animals and humans. The toxicology database for mesotrione is not complete. The HIARC recommended that a developmental neurotoxicity study be required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects. The HIARC also requested a 28-day inhalation study to characterize the direct effects of mesotrione on the pulmonary system and systemic effects via the inhalation route. Mesotrione has low acute toxicity via the oral, dermal, and inhalation routes. It is a mild eye irritant, but is not a dermal irritant or a dermal sensitizer.

The eye, liver, and kidney are the primary target organs of mesotrione. Ocular effects such as corneal opacity, corneal vascularization, and keratitis were observed in rat in subchronic, chronic, and reproduction studies and in the dog in a chronic study. Lenticular opacity was also noted in the chronic dog study. Ocular effects were observed in mice in the reproduction study. Liver effects included increased liver weights seen in the rat subchronic, chronic, and reproduction studies and hepatocyte fat vacuolation noted in the rat chronic study. Kidney effects included increased kidney weights observed in the rat subchronic and chronic studies and in the rat and mouse reproduction studies and hydronephrosis noted in the rat subchronic and reproduction studies. Body weight decrements and/or decreased food efficiency were noted in the mouse chronic and carcinogenicity studies and the rat developmental study.

Plasma tyrosine levels were increased in the rat, mouse and dog in the chronic and reproduction studies in which levels were measured. The ocular, liver and kidney effects have been demonstrated to be mediated by the high tyrosine levels in the blood caused by inhibition of the enzyme HPPD. The rat is the most sensitive species to this effect compared to the dog and the mouse. The Mechanism of Toxicity Science Assessment Review Committee determined that for tyrosine-mediated toxicological effects, the mouse is a more appropriate model for assessing human risk than is the rat. This decision was based on comparative data on the activity of tyrosine aminotransferase (TAT) activity in the rat, mouse and human, and the similarities of the response to elevated plasma tyrosine levels in humans and the mouse (D272633; March 27, 2001).

Long-term dietary administration of mesotrione did not result in an overall treatment-related increase in incidence of tumor formation in rats or mice. The HIARC classified mesotrione as "not likely to be carcinogenic to humans" by all routes of exposure based upon lack of evidence of carcinogenicity in rats and mice.

Mesotrione did not show evidence of mutagenicity in *in vitro* or *in vivo* studies.

Oral rat, mouse, and rabbit developmental studies showed an increased susceptibility of the fetus to mesotrione *in utero*. Delayed ossification was seen in the fetuses at doses below those at

7

which maternal toxic effects were noted. Maternal toxic effects in the rat were decreased body weight gain during treatment and decreased food consumption and in the rabbit, abortions and GI effects. No maternal toxic effects were noted in the mouse. Multi-generation reproduction studies also showed an increased susceptibility of the young to mesotrione. In the mouse, the young exhibited significant tyrosinemia and ocular discharge at doses below those at which parental toxic effects were noted. The parental effects were tyrosinemia and increased kidney weights. In the rat, no NOAEL was determined for parental effects (tyrosinemia, increased liver weights) or offspring systemic effects (tyrosinemia); however, the tyrosinemia was much more severe in the young than in the adults. Decreased litter size was noted at the next highest dose.

No evidence of neurotoxicity or neuropathology was seen in the acute and subchronic neurotoxicity studies. In the multi-generation mouse reproduction study, one F_1 male and one F_1 female had retinal detachment with marked cataractous changes at the highest dose tested (>1000 mg/kg/day). In the subchronic toxicity dog study, the high-dose females had decreased absolute and relative brain weights; however, no microscopic abnormalities were noted in any brain tissue from the high-dose group and the effect was not observed in the chronic toxicity dog study. There is some concern about the effects of elevated plasma tyrosine levels on the developing nervous system in children due to a report by Ruetschi *et al* (2000)[3] that some patients with tyrosinemia III (an autosomal recessive disorder in which 4-hydroxyphenylpyruvate dioxygenase is deficient, resulting in high plasma tyrosine levels) were presented with mental retardation or neurological symptoms and that no correlation of the severity of the mutation and enzyme deficiency and mental function has been found. Also, tyrosine levels did not correlate with the clinical phenotype.

A series of rat metabolism studies with [^{14}C-aromatic] mesotrione indicated that mesotrione was readily absorbed and distributed in the body. Tissue distribution was about the same in both sexes, although one study showed higher residues in the kidneys in females, with the highest residues of the test compound in the liver and kidney. Higher doses resulted in higher residues in the liver and kidney, while repeated doses resulted in reduced accumulation of residues in all tissues. Levels of radioactivity in tissues of iv-dosed animals were essentially the same as in orally-dosed animals. Over 50% of the administered dose was excreted in the urine in both sexes and around 25% was excreted in the feces within 72 hours. Females exhibited slightly higher total urinary excretion than males, but total fecal excretion was about the same in both sexes. Increasing the dose or repeated doses had little effect on the pattern of excretion in both sexes. The overall pattern of excretion was similar between orally-dosed and iv-dosed rats. The metabolite profile was similar between the sexes in each group and between the single-dosed and repeated-dosed animals. The parent compound, mesotrione, was the major component identified in the urine accounting for 47-64% of the dose. In addition, the following minor metabolites were identified: MNBA (1-4% of the dose), AMBA (3-12%), 5-hydroxymesotrione ($\leq$ 2%), and

---

[3]Ruetschi, U., *et al.*, (2000) Mutations in the 4-hydroxyphenylpyruvate dioxygenase gene (HPD) in patients with tyrosinemia type III. HUM. Genet. 106(6): 654-662.

8

4-hydroxymesotrione (3-6%). In bile cannulated rats administered [^{14}C-aromatic]mesotrione or [^{14}C-dione]mesotrione, the major component in fecal excreta and bile was the parent compound. Analysis of the bile identified mesotrione and 4-hydroxymesotrione as two minor components. Another minor component in the feces was 5-hydroxymesotrione. Metabolism in the mouse was very similar to the rat except that males had slightly increased total fecal excretion when compared to females and, females in the low-dose group excreted higher (1.5x) levels of parent compound in the urine than males. Free mesotrione was the major component in the urine and feces ($\geq$ 50% of the dose). Minor components in the fecal extracts included AMBA (1-4%) and MNBA ($\leq$ 2%).

The mesotrione metabolites, MNBA and AMBA, have low acute oral toxicity and MNBA has low dermal toxicity. Reverse gene mutation assays indicated that MNBA and AMBA are not mutagenic; liver cytosol studies indicated that these metabolites are weak inhibitors of HPPD *in vitro*. MNBA, when administered to adult rats by gavage at up to 1000 mg/kg/day for 28 days, caused an increase in motor activity in females, but no other signs of neurotoxicity were observed.

2.0 REQUIREMENTS

The requirements (CFR 158.340, revised as of July 1, 1999) for Food Use for mesotrione technical are in Table 1. Use of the new guideline numbers does not imply that the new (1998) guideline protocols were used.

Table 1.

Test		Technical	
		Required	Satisfied
870.1100	Acute Oral Toxicity	yes	yes
870.1200	Acute Dermal Toxicity	yes	yes
870.1300	Acute Inhalation Toxicity	yes	yes
870.2400	Primary Eye Irritation	yes	yes
870.2500	Primary Dermal Irritation	yes	yes
870.2600	Dermal Sensitization	yes	yes
870.3100	Oral Subchronic (Rodent)	yes	yes
870.3150	Oral Subchronic (Non-Rodent)	yes	yes
870.3200	21-Day Dermal	yes	yes
870.3250	90-Day Dermal	no	-
870.3465	90-Day Inhalation	yes[a]	no
870.3700a	Developmental Toxicity (Rodent)	yes	yes
870.3700b	Developmental Toxicity(Non-rodent)	yes	yes
870.3800	Reproduction	yes	yes

Table continues next page

Test		Technical	
		Required	Satisfied
870.4100a	Chronic Toxicity (Rodent)	yes	yes
870.4100b	Chronic Toxicity (Non-rodent)	yes	yes
870.4200a	Oncogenicity (Rat)	yes	yes[b]
870.4200b	Oncogenicity (Mouse)	yes	yes
870.4300	Chronic/Oncogenicity	yes	yes
870.5100	Mutagenicity—Gene Mutation - bacterial	yes	yes
870.5300	Mutagenicity—Gene Mutation - mammalian	yes	yes
870.5375	Mutagenicity—Structural Chromosomal Aberrations	yes	yes
870.5395	Mutagenicity—Other Genotoxic Effects	no	-
870.6100a	Acute Delayed Neurotox. (Hen)	no	-
870.6100b	90-Day Neurotoxicity (Hen)	no	-
870.6200a	Acute Neurotox. Screening Battery (Rat)	yes	yes
870.6200b	90 Day Neuro. Screening Battery (Rat)	yes	yes
870.6300	Developmental. Neurotoxicity	yes[c]	no
870.7485	General Metabolism	yes	yes
870.7600	Dermal Penetration	no	-
Special Studies for Ocular Effects			
	Acute Oral (Rat)	no	-
	Subchronic Oral (Rat)	yes	yes
	Six-month Oral (Dog)	no	-

a The HIARC recommended that a 28-day inhalation study be required.
b The 2-year feeding study in rats is used to satisfy the data requirements for 870.4200.
c The HIARC recommended that a developmental neurotoxicity study be required.

3.0 DATA GAP(S)

The toxicology database for mesotrione is not complete. The HIARC recommended that a developmental neurotoxicity study be required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects. The HIARC also requested a 28-day inhalation study to characterize the direct effects of mesotrione on the pulmonary system and systemic effects via the inhalation route.

4.0 HAZARD ASSESSMENT

4.1 Acute Toxicity

Adequacy of data base for acute toxicity: The data base for acute toxicity is considered complete. No additional studies are required at this time. Mesotrione and its SC formulation

10

have low acute toxicity via the oral, dermal, and inhalation routes. The mesotrione metabolites, MNBA and AMBA, have low acute oral toxicity and MNBA has low dermal toxicity. Mesotrione technical is a mild eye irritant, but is not a dermal irritant or a dermal sensitizer. The SC formulation is a slight eye and dermal irritant and a mild dermal sensitizer. Acute toxicity data for mesotrione technical and its metabolites and the SC formulation are summarized below in Tables 2a and 2b.

Table 2a. Acute Toxicity Data on Mesotrione and its MNBA and AMBA Metabolites

Guideline No./Study Type		MRIDs	Results	Tox Category
870.1100	Acute Oral	44373512 44505015[a] 44505016[b]	LD_{50} > 5000 mg/kg	IV
870.1200	Acute Dermal	44373514 45196004[a]	LD_{50} > 2000 mg/kg	III
870.1300	Acute Inhalation	44373516	LC_{50} > 4.75 mg/L	IV
870.2400	Primary Eye Irritation	44373518	Mild eye irritant	IV
870.2500	Primary Skin Irritation	44373520	Not a dermal irritant	IV
870.2600	Dermal Sensitization	44373522	Not a dermal sensitizer	N/A

a MNBA metabolite
b AMBA metabolite

Table 2b. Acute Toxicity Data on Mesotrione 480 g/L SC [40.5% (w:w) ai] Formulation

Guideline No./Study Type		MRIDs	Results	Tox Category
870.1100	Acute Oral	44373513	LD_{50} > 5000 mg/kg	IV
870.1200	Acute Dermal	44373515	LD_{50} > 2000 mg/kg	III
870.1300	Acute Inhalation	44373517	LC_{50} > 2.83 mg/L	IV
870.2400	Primary Eye Irritation	44373519	Slight eye irritant	IV
870.2500	Primary Skin Irritation	44373521	Slight dermal irritant	IV
870.2600	Dermal Sensitization	44373523	Mild dermal sensitizer	N/A

4.2 Subchronic Toxicity

Adequacy of data base for subchronic toxicity: The data base for subchronic toxicity is not complete. There are no appropriate inhalation studies, and the HIARC requested a 28-day inhalation study to characterize the direct effects of mesotrione on the pulmonary system and systemic effects via the inhalation route. The eyes were the primary target organ of mesotrione in the subchronic rat studies. Ocular effects such as corneal opacity, corneal vascularization, and keratitis as well as increased liver weights were observed in both sexes. In males, overall decreased body weight gains and increased kidney weights were observed at the LOAEL in one study and minimal to slight unilateral hydronephrosis and minimal chronic progressive glomerulonephropathy were observed at >LOAEL in a subsequent study. No adverse effects were observed in dogs or mice treated orally and rabbits treated dermally with mesotrione up to the limit dose. Special studies in rodents showed that treatment with mesotrione caused tyrosinemia with associated increases in ocular lesions and urinary phenolic acids output, changes in liver enzyme activities, and adaptive changes in liver in rats and mice and kidney weights in rats. Also, treatment with mesotrione resulted in minimal CYP induction in rats and mice. Mesotrione in combination with tyrosine in the diet caused marked tyrosinemia and associated ocular lesions and changes in liver enzyme activities in rats. In general, treatment with the combination caused more marked effects than treatment with either compound alone. Other special studies with the mesotrione metabolites MNBA and AMBA, resulted in weak inhibition of rat HPPD *in vitro* and MNBA, administered to adult rats by gavage at up to 1000 mg/kg/day for 28 days caused increased motor activity in the females. Special studies with mesotrione in humans indicated that oral dosing with mesotrione elevated plasma tyrosine levels and dermally applied mesotrione appeared to remain in the stratum corneum and had no discernable effect on plasma tyrosine levels.

870.3100 90-Day Oral Toxicity - Rodent

1) In this subchronic oral toxicity study (MRID 44505019), mesotrione (93.3% a.i., Lot/batch # P8) was administered for 90 days to 12 Alpk:AP$_f$SD rats/sex/dose at dietary concentrations of 0, 1, 125, 1250, or 12500 ppm (equivalent to [M/F] 0/0, 0.09/0.10, 11/13, 112/126, and 1111/1213 mg/kg/day, respectively).

No treatment-related findings were observed in the 1 ppm group. No mortalities occurred during the study. Hematology, clinical chemistry, and urinalysis parameters were unaffected by the test substance.

The eye was the main target organ. At the clinical, ophthalmoscopic, and gross pathological examinations, corneal lesions (eye opacity and vascularization) were observed in both sexes of the 125, 1250, and 12500 ppm dose groups. Upon histological examination, the corneal lesions were characterized as keratitis.

12

In the 125 ppm males, mean body weights (adjusted for week 1 body weight) were decreased ($p \leq 0.01$ or 0.05) sporadically throughout the first half of the study (↓2-5%) and from week 10 through study termination (↓6-8%). Body weights in the 125 and 1250 ppm females were also decreased ($p \leq 0.05$) sporadically throughout the study. Overall (weeks 1-14) body weight gains were decreased in both sexes of the 125, 1250, and 12500 ppm dose groups (↓4-23%, calculated by reviewers). Biological significance was noted at ≥ 125 ppm in males and at 12500 in females. Overall food utilization (weeks 1-13) was decreased ($p \leq 0.01$) in the males (↓7-13%) at ≥ 125 ppm, but was only considered biologically relevant at 1250 and 12500 ppm.

Additionally in the 12500 ppm group, decreased ($p \leq 0.01$ or 0.05) mean body weights (↓6-16%) and food consumption (↓9-18%) were observed throughout the study.

In the 125, 1250, and 12500 ppm animals, increased ($p \leq 0.01$ or 0.05) adjusted (for final body weight) liver (↑11-19%) and kidney (↑10-14%, males only) weights were observed. There were no corroborative histological findings to indicate an adverse effect on these organs.

The LOAEL for this study is 125 ppm (equivalent to 11 mg/kg/day in males and 13 mg/kg/day in females) based on corneal abnormalities observed during the clinical, ophthalmoscopic, gross pathological, and histopathological examinations in both sexes, and decreases in body weight gain in males.

The NOAEL for this study is 1 ppm (equivalent to 0.09 mg/kg/day in males and 0.10 mg/kg/day in females).

The submitted study is classified as **acceptable/guideline (§82-1a)** and satisfies the requirements for a subchronic oral toxicity study in rats.

2) In this subchronic oral toxicity study (MRID 44505020), mesotrione (96.8% a.i., Lot/batch # P17) was administered for 90 days to 12 Alpk:AP$_f$SD rats/sex/dose at dietary concentrations of 0, 2.5, 5.0, 7.5, or 150 ppm (equivalent to [M/F] 0/0, 0.21/0.23, 0.41/0.47, 0.63/0.71, or 12.46/14.48 mg/kg/day, respectively).

No treatment-related findings were observed in the 2.5 or 5.0 ppm groups. No mortalities occurred during the study. Body weights (adjusted for week 1 body weight), body weight gains, food consumption and utilization, hematology, clinical chemistry, and urinalysis parameters, and organ weights were unaffected by the test substance.

Cloudy eyes were observed during the clinical examinations during weeks 8-14 in the 7.5 ppm males and during weeks 7-14 in the 150 ppm males. In the 150 ppm females, cloudy eyes were observed during week 12 only. During the ophthalmoscopic examination at week 13, slight to marked hazy opacity, slight to moderate opacity, and vascularization were observed in the 7.5

B

and 150 ppm males. Slight opacity was observed in the 150 ppm females and 5 ppm males. In addition, plaque opacity of the lens was observed in the 150 ppm females. Eye opacity was observed in the 7.5 and 150 ppm males during the gross pathological examination. In addition, slight to moderate keratitis of the eye was observed in the 7.5 and 150 ppm males and in the 150 ppm females during the histopathological examination. Histopathological abnormalities of the kidney included minimal to slight unilateral hydronephrosis and minimal chronic progressive glomerulonephropathy in the 150 ppm males.

The LOAEL for this study is 7.5 ppm (equivalent to 0.63 mg/kg/day for males, 0.71 mg/kg/day for females) based upon corneal lesions in males. The NOAEL is 5 ppm (equivalent to 0.41 mg/kg/day for males, 0.47 mg/kg/day for females).

The submitted study is classified as **acceptable/guideline (§82-1a)** and satisfies the requirements for a subchronic oral toxicity study in rats.

3) In this subchronic oral toxicity study (MRID 44505022), mesotrione (ZA 1296; 96.8% a.i.; Batch No. P17) was administered for 13 weeks to 20 C57BL/10J$_f$CD-1 mice/sex/dose at dietary concentrations of 10, 50, 350, or 7000 ppm (equivalent to 1.7, 8.4, 61.5, and 1212.4 mg/kg/day in males and 2.4, 12.4, 80.1, and 1537.1 mg/kg/day in females). Two control groups of 20 C57BL/10J$_f$CD-1 mice/sex (40/sex) were fed untreated diet during the study.

Mortality, clinical observations, body weight, body weight gain, food consumption and utilization, ophthalmoscopic observations, hematology and clinical chemistry parameters, organ weights, and gross and microscopic pathological findings were unaffected by the test substance.

The NOAEL for this study is 7000 ppm (equivalent to 1212/1537 mg/kg/day [M/F]). The LOAEL was not observed.

The submitted study is classified as **acceptable/guideline (§82-1)** and satisfies the requirements for a subchronic oral toxicity study in rodents. Although a LOAEL was not observed, the study was tested up to the limit dose.

870.3150 90-Day Oral Toxicity - (Non-Rodent)

In this subchronic oral toxicity study (MRID 44505023), mesotrione (ZA1296; 96.8% a.i., Lot/batch # P17) was administered via gelatin capsule for 90 days to 4 Beagle dogs/sex/dose at dietary concentrations of 0, 100, 600, or 1000 mg/kg/day.

No mortalities occurred during the study. Clinical signs, body weight, food consumption, hematology and clinical chemistry parameters, organ weights, and gross and microscopic pathological findings were not adversely affected by the test substance. Absolute brain weight and adjusted (for body weight) brain weight were decreased in the high-dose females (↓12%

each, $p \leq 0.05$); however, no microscopic abnormalities were noted in any brain tissue from the high-dose group.

The incidence of reddened ears increased at 600 (1/4 males, 2/4 females) and 1000 (3/4 males, 3/4 females) mg/kg in both sexes as compared to the controls (0/4 males, 1/4 females). However, since no associated adverse effects were noted with this clinical observation, it is not considered toxicologically relevant.

The NOAEL for this study is 1000 mg/kg/day based on the lack of any adverse effects up to the limit dose. The LOAEL is > 1000 mg/kg/day.

The submitted study is classified as **acceptable/guideline (§82-1b)** and satisfies the requirements for a subchronic oral toxicity study in dogs.

870.3200 21-Day Dermal Toxicity -Rabbit

In a repeated-dose dermal toxicity study (MRID 44505024), mesotrione (96.8% purity, Lot/Batch #: P17 [WRC 15213-17-1]) was applied to the clipped intact skin of five New Zealand White albino rabbits/sex/dose at nominal doses of 0 (vehicle control), 10, 500, or 1,000 mg/kg/day (limit dose) for 6 hours/day, 5 days/week, for a total of 15 applications during a 21-day period.

No rabbits died during the course of the study as a result of treatment and no treatment-related clinical signs of systemic toxicity were observed. Slight erythema was observed in 4/10 mid-dose animals and 10/10 high-dose animals; however, all irritation subsided by 22 days (study termination) and no gross or microscopic treatment-related dermal abnormalities were observed upon necropsy. No other treatment-related differences in toxicity, body weights, food consumption, hematology, ophthalmology, clinical chemistry, organ weights, or gross or microscopic pathology were observed between the control and treated groups.

The LOAEL was not established.
The NOAEL is ≥1,000 mg/kg/day (limit dose).

This study is classified **acceptable/guideline (§82-2)** and does satisfy the requirement for a repeated-dose dermal toxicity study. Although a LOAEL was not observed, the test substance was tested up to the limit dose.

870.3250 90-Day Dermal

There are no 90-day dermal studies.

870.3465 90-Day Inhalation – Rats

There are no appropriate inhalation studies. The HIARC requested a 28-day inhalation study to characterize the direct effects of mesotrione on the pulmonary system and systemic effects via the inhalation route.

Special Studies - Rodent

1) The stated objective of this study (MRID 44537103) was to investigate the reversibility of liver and kidney weight changes in rats induced by dietary administration of mesotrione for 90 consecutive days. Tyrosinemia occurs in animals fed mesotrione or other triketones due to an inhibition of the liver enzyme p-hydroxyphenylpyruvate dioxygenase (HPPD). HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. HPPA is formed from tyrosine by the action of the liver enzyme tyrosine aminotransferase (TAT). Liver TAT activity can be elevated by high serum tyrosine levels. Male Alpk:ApfSD rats (40/group with 2 control groups) were fed diets containing mesotrione (96.8% purity, batch P17) at 0, 5, 100, or 2500 ppm (equivalent to 0, 0.37, 7.52, and 192 mg/kg/day, respectively) for 90 days. Eight rats/group were terminated at 90 days, 1 week recovery (2500 ppm and 1 control group, only), 2 weeks recovery, 4 weeks recovery, 6 weeks recovery (5 and 100 ppm and 1 control group, only), and 9 weeks recovery. Clinical signs, body weights, food consumption, plasma tyrosine levels, kidney and liver weights, and liver TAT and HPPD levels were measured and/or recorded. Liver and kidney tyrosine levels were determined for the 5 and 100 ppm groups. Liver and kidney tissue samples from the 2500 ppm group and one control group were examined by light (all animals) and electron (4/group) microscopy. Ophthalmoscopic exams were performed at the end of the 90-day treatment period and during the recovery period prior to termination. Necropsies were performed at termination or upon the event of premature death.

The results of this special study are presented as attachments to this overview (Study Report Tables 3, 4, 5, and 7 through 12, pages 41, 42, 45 through 47, 49 through 51, 61 through 68, 71, 73, 76, and 78 through 82). There were no differences of toxicological concern in food consumption. No treatment-related observations of toxicological concern were made at histopathological or electron microscopic examination. No treatment-related mortalities occurred.

Adjusted (to week 1) body weights were decreased in 2500 ppm rats from weeks 3-21 of treatment (↓2-10%, $p \leq 0.05$ or 0.01). Food consumption was slightly decreased in 2500 ppm rats from weeks 5-8 and 10-13 (↓4-7%, $p \leq 0.05$ or 0.01). Decreases ($p \leq 0.05$ or 0.01) of toxicological concern in food utilization occurred in the 2500 ppm rats during the weeks 5-8 (↓9%), 9-13 (↓20%), and 1-13 (↓9%), and in the 100 ppm rats during weeks 9-13 (↓9%). During clinical examination, cloudy/opaque eyes were observed in the 5, 100, and 2500 ppm groups (1, 37, and 33/40 treated animals, respectively, vs 0 controls) during the treatment period. The opacity resolved during the recovery period, and no observations were made from week 17 onwards. It

was stated that the only ophthalmoscopic finding in controls was a slight, unilateral haziness at 9 weeks of recovery; no data were provided. At ophthalmoscopic exam, the incidence of slight to marked hazy opacity or slight to marked opacity was increased in the 5, 100, and 2500 ppm animals at 90 days of treatment (10/80, 56/80, and 46/76 eyes examined, respectively, vs 0 in controls). At 1 (2500 ppm) or 2 (5 ppm) weeks of recovery, ocular opacity had completely resolved in the 5 and 2500 ppm treatment groups. One 2500 ppm animal did have slight hazy opacity after 4 weeks recovery. In the 100 ppm animals, opacity was still apparent during the recovery period at 2 weeks (4/16 eyes, slight hazy opacity), 4 weeks (8/16, minimal hazy opacity), and 6 weeks (4/16, slight hazy opacity). Corneal vascularization was observed in 5, 100, and 2500 ppm animals (2/80, 44/80, and 41/76 eyes examined, respectively, vs 0 in controls) at the end of the treatment period and had resolved by the next examination (1 or 2 weeks of recovery). Ghost vascularization of the cornea was observed during recovery in the 100 ppm group at weeks 2, 6, and 9 (10/16, 14/16, and 4/16 eyes examined, respectively) and in the 2500 ppm group at weeks 1, 2, 4, and 9 (8/14, 10/16, 8/16, and 10/14 eyes examined, respectively). Ghost vascularization was observed in one 5 ppm animal at 6 weeks recovery.

Plasma tyrosine concentrations were elevated ($p \leq 0.01$) with respect to controls at the first timepoint in the 5 (1 week), 100 (1 week) and 2500 (24 hours) ppm group (↑818%, 1460%, and 2215%, respectively) and at the end of treatment (week 14 -↑726, 1279, and 939%, respectively). Plasma tyrosine remained elevated ($p \leq 0.01$) during the recovery period in the 2500 ppm group at weeks 15 (↑240%), 16 (↑274%), and 23 (↑21%). Plasma tyrosine levels remained slightly elevated ($p \leq 0.05$ or 0.01) in the 5 and 100 ppm groups at weeks 16 (↑22-45%) and 20 (↑17-22%). Kidney tyrosine levels were increased in the 5 and 100 ppm groups at the end of treatment (week 14 - ↑246 and 402%, respectively; $p \leq 0.05$). Liver tyrosine concentration was also increased in both groups at the end of treatment (↑626 and 1102%, respectively; $p \leq 0.01$). Levels had returned to normal by week 16. TAT activity was increased ($p \leq 0.01$ in 2500 ppm group only) in all treatment groups at the end of treatment (week 14 - ↑15-66%). At week 15, TAT activity was slightly elevated in the 2500 ppm group (↑28%, p = not significant). By week 16, TAT levels were not different from controls in any treatment group. HPPD activity was decreased ($p \leq 0.01$) at the end of treatment in all treatment groups (↓89-96%). During the recovery period, HPPD activity slowly returned to control levels, but was still depressed at week 23 (↓22-36%; $p \leq 0.01$ in 5 and 100 ppm groups, not significant in 2500 ppm group). Adjusted (to body) liver weights were increased dose-dependently at week 14 in all treatment groups (↑10-20%, $p \leq 0.01$). At subsequent timepoints (weeks 15, 16, 18, and 23), adjusted liver weights were consistently increased only in the 2500 ppm group (↑10-14%; $p \leq 0.05$ or 0.01). Absolute liver weights were increased ($p \leq 0.05$ or 0.01) in the 5 and 100 ppm groups at week 14 (↑11%), but not in the 2500 ppm group. Absolute liver weights were not increased at later dates except in the 5 ppm group at week 23 (↑12%, $p \leq 0.05$). Adjusted (to body) kidney weights were increased ($p \leq 0.05$ or 0.01) at week 14 in 5, 100, and 2500 ppm groups (↑12, 12, and 10%, respectively), whereas absolute kidney weights were increased ($p \leq 0.05$ or 0.01) in 5 and 100 ppm groups only (↑13 and 11%, respectively). Increases ($p \leq 0.05$ or 0.01) in adjusted kidney weights also occurred in the 2500 ppm group at week 15 (↑5%) and week 23 (↑15%). Increases in absolute kidney weights at later dates occurred only in the 5 ppm group at week 23 (↑12%, $p \leq 0.05$).

Decreases in adjusted (to body) and absolute brain weights occurred only in the 100 ppm group at week 14 (↓4%, $p \leq 0.05$)

In conclusion, treatment with mesotrione caused tyrosinemia with associated increases in ocular lesions, changes in liver enzyme activities, and adaptive changes in liver and kidney weights. Upon removal of mesotrione from the diet, complete recovery was observed in the ocular lesions, plasma, liver, and kidney tyrosine levels, TAT activity, and kidney weights. Partial recovery was observed in HPPD activity and liver weights.

2) The stated objective of this study (MRID 44537106) was to investigate the correlation between mesotrione-induced tyrosinemia and ocular, body weight, and organ weight changes in male Alpk:Ap$_f$SD rats. Tyrosinemia occurs in animals fed mesotrione or other triketones due to an inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD). HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. HPPA is formed from tyrosine by the action of tyrosine aminotransferase (TAT). Male Alpk:Ap$_f$SD rats (16/group with 2 control groups) were fed diets containing mesotrione (96.8% purity, batch P17) at 0, 0.5, 1, 3, 4, 5, 7.5, 10, or 100 ppm (equivalent to 0, 0.04, 0.09, 0.27, 0.35, 0.44, 0.67, 0.89, and 8.96 mg/kg/day) for 90 days. Clinical signs, body weights, food consumption, plasma tyrosine levels, urine phenolic acid levels, kidney and liver weights, and liver TAT and HPPD levels were measured and/or recorded. Liver and kidney tissue samples were examined by light and electron microscopy. Ophthalmoscopic exams were performed during the 1 to 2-week acclimatization period and during the last week of the study. Necropsies were performed at termination or upon the event of premature death.

The results of this special study are presented as attachments to this overview (Study Report Tables 4, 5, 7 through 10, 13, and 14, pages 40, 42 through 45, 50 through 54, 60, and 61). There were no differences of toxicological concern in body weights, body weight gains (as calculated by the reviewers), or food utilization. There were no differences in food consumption. No treatment-related observations were made at histopathological or electron microscopic examination.

Cloudy/opaque eyes were observed during clinical examinations in the 5, 7.5, 10, and 100 ppm groups (2/16, 1/16, 3/16, and 13/16 treated animals, respectively, vs 0/32 controls). At ophthalmoscopic exam, the incidence of slight to moderate hazy opacity or slight to moderate opacity was increased in the 7.5, 10, and 100 ppm animals (5/32, 6/32, and 22/32 eyes examined, respectively, vs 1/64 controls). Corneal vascularization was observed in 7.5, 10, and 100 ppm animals (1/32, 2/32, and 15/32 eyes examined, respectively, vs 0/64 controls). Ghost vascularization of the cornea was observed in one 100 ppm male only (1/32 eyes examined vs 0/64 controls). At necropsy, opaque eyes were observed in the 7.5, 10, and 100 ppm groups (1/16, 3/16, and 7/16 animals treated, respectively, vs 0/32 controls). Total urinary phenolic acids were increased in all treatment groups at week 13 (↑114-1247%), and reflect the increase in

both conjugated (↑53-472%) and free (↑130-2121%) phenolic acids. The proportion of conjugated to free phenolic acids tended to decrease with increasing dose (1.13-0.17 [conjugated/free]), although not strictly dose-dependently. Plasma tyrosine concentrations were elevated ($p \leq 0.01$) with respect to controls in a dose-dependent manner at 24 hours in the 1 - 100 ppm groups (↑40-2661%) and in all treatment groups at weeks 1 (↑292-2696%) and 14 (↑102-2353%). TAT activity was increased ($p \leq 0.05$ or 0.01) at termination with respect to controls in the 3-100 ppm groups (↑35-61%), and HPPD activity was decreased ($p \leq 0.01$) at termination with respect to controls in all treatment groups (↓68-97%), although neither changes were dose-dependent. Relative liver weights were increased ($p \leq 0.05$ or 0.01) in a dose-dependent manner in the 4-100 ppm groups (↑5-15%), whereas absolute liver weights were increased ($p \leq 0.05$ or 0.01) in the 5-100 ppm groups (↑8-13%), but not dose-dependently. Relative kidney weights were increased ($p \leq 0.05$ or 0.01) in 5, 10, and 100 ppm groups (↑4-8%) and absolute kidney weights were increased ($p \leq 0.05$ or 0.01) in 5 and 10 ppm groups only (↑9-10%). There were no treatment-related observations made in the liver or kidneys at histopathological or electron microscopic examination to suggest that the changes in organ weights were other than an adaptive response.

In conclusion, treatment with increasing levels of mesotrione caused a dose-dependent tyrosinemia with associated increases in ocular lesions and urinary phenolic acids output, changes in liver enzyme activities, and adaptive changes in liver and kidney weights.

3) The objective of this study (MRID 44537107) was to investigate the correlation between mesotrione-induced tyrosinemia and ocular, body weight, and organ weight changes in female Alpk:Ap$_f$SD rats. Tyrosinemia occurs in animals fed mesotrione or other triketones due to an inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD). HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. HPPA is formed from tyrosine by the action of tyrosine aminotransferase (TAT). Female Alpk:Ap$_f$SD rats (20/group) were fed diets containing mesotrione (96.8% purity, Batch P17) at 0, 1, 5, 10, 50, 100, 1000, or 2500 ppm (equivalent to 0, 0.09, 0.48, 0.95, 4.82, 9.54, 94.83, and 236.75 mg/kg/day) for 90 days. Four animals/group were sacrificed at 7 and 29 days; the remainder (12/group) were sacrificed at 90 days.

Clinical signs, body weights, food consumption, plasma tyrosine levels, urine ketone levels, kidney and liver weights, and liver TAT and HPPD levels were measured or recorded. Ophthalmoscopic exams were performed during the last week of the study. Necropsies were performed at termination or upon the event of premature death.

Results of toxicological concern of this special study are presented as an attachment to this overview (Study Report Tables 3, 4, 5, and 8 through 12, pages 34, 38, 42, 43, 49, 50, 52, 55, 56, 57, and 58). There were no differences of toxicological concern in food consumption, food utilization, or kidney weights. Gross necropsy results were not reported.

Body weights were decreased in the 2500 ppm group from week 4 to 14, although not continuously (↓4-6%, $p \leq 0.05$ or 0.01). Body weight gain (as calculated by the reviewers) was decreased in the 2500 ppm group (↓9%). Cloudy eyes were observed during clinical examinations in the 1000 and 2500 ppm groups (7 animals in each treated group vs 0 controls). At ophthalmoscopic exam, the incidence of slight to moderate hazy opacity or slight to moderate opacity was increased in the 100, 1000, and 2500 ppm animals (2/24, 16/24, and 15/24 eyes examined, respectively, vs 1/24 controls). Corneal vascularization was observed in 1000 and 2500 ppm animals (3/24 and 2/24 eyes examined, respectively, vs 0/24 controls). Ghost vascularization of the cornea was observed in one 1000 and one 2500 ppm female (1/24 eyes examined each vs 0/24 controls). Total urinary phenolic acids were increased at week 5 in 100, 1000, and 2500 ppm treatment groups (1.82, 6.95, and 23.0 mg equivalents, respectively, vs not detected in controls). The proportion of conjugated to free phenolic acids tended to decrease with increasing dose (100% conjugated in the 100 ppm group, 18% conjugated in 1000 ppm group, and 12% conjugated in 2500 ppm group). Plasma tyrosine concentrations were elevated ($p \leq 0.01$) with respect to controls in a dose-dependent manner in the 5 - 2500 ppm groups at weeks 2 (↑94-1389%), 5 (↑77-1123%), and 14 (↑72-1154%; Note - 1000 ppm change [1154%] slightly exceeded 2500 ppm change [1108%]). TAT activity was increased ($p \leq 0.05$ or 0.01) with respect to controls at week 2 in the 5-2500 ppm groups (↑65-153%), and at week 5 (↑87-113%) and 14 (↑27-43%) in the 1000 and 2500 ppm groups. HPPD activity was decreased ($p \leq 0.01$) with respect to controls in all treatment groups at week 2 (↓56-99%), week 5 (↓71-98%), and week 14 (↓60-99%). The effect plateaued at ≥5 ppm. Adjusted (to body) liver weights were increased ($p \leq 0.05$ or 0.01) at week 5 in the 5, 50, 100, and 2500 ppm groups (↑6-14%) and at week 14 in the 1000 and 2500 ppm groups (↑6-7%), whereas absolute liver weights were increased ($p \leq 0.01$) in the 50, 100, and 2500 ppm groups at week 5 only (↑19-22%). The increases were not dose-dependent.

In conclusion, treatment with increasing levels of mesotrione caused a dose-dependent tyrosinemia with associated increases in ocular lesions and urinary phenolic acids output, changes in liver enzyme activities, and adaptive changes in liver weights.

4) The objective of this study (MRID 44505111) was to investigate the effect of mesotrione (96.8% purity, Batch P17) on tyrosinemia in female Alpk:Ap$_f$SD rats given a high-tyrosine diet. Tyrosinemia occurs in animals fed mesotrione or other triketones due to an inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD). HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. HPPA is formed from tyrosine by the action of tyrosine aminotransferase (TAT). Female Alpk:Ap$_f$SD rats (8/treatment group) were fed diet with 0, 0.5, 1.0, or 2.5% tyrosine or 100 ppm mesotrione diet with 0, 0.5, 1.0, or 2.5% tyrosine for 28 days. Body weights, food consumption, plasma tyrosine levels, urine ketone body levels, organ weights, and liver TAT and HPPD levels were measured. Ophthalmoscopic exams and necropsies were also performed.

The results of this special study are presented as an attachment to this overview (Study Report Tables 2 through 9, pages 26 through 33). Cloudy eyes were observed at clinical examinations in 8/8 animals in the 3 mesotrione/tyrosine-treated groups, only. Body weights (↓9-11%) and body weight gains (↓23-31%) were reduced ($p \leq 0.01$) with respect to controls in animals receiving 100 ppm mesotrione/2.5% tyrosine (100 ppm/2.5% Tyr), only. Decreased food consumption was observed in the 100 ppm/2.5% Tyr animals (↓8-17%) throughout the study. Food utilization (bodyweight gain [g]/100 g food consumed) was not significantly different between treated and control groups. At the ophthalmoscopic exam, minimal to marked corneal opacity was observed in all mesotrione-treated groups, the effect becoming more pronounced with increasing tyrosine levels (1/16 - 16/16 eyes treated vs 0/16 controls). Corneal vascularization was observed in the 100 ppm/1.0% Tyr (10/16 eyes) and 100 ppm/2.5% Tyr (16/16 eyes) treatment groups (vs 0/16 control eyes). Plasma tyrosine concentrations were elevated with respect to controls at all timepoints in the 2.5% Tyr groups (↑73-139%) and in a dose-dependent manner in all mesotrione-treated groups (24 hours post-dose -↑1236-3026%; 1 week post-dose -↑778-2535%; termination - ↑1012-2522%). Urine ketone level were not reported. It was stated that no acetoacetate was detected in any of the urines analyzed, but that HPPA was probably present in high concentrations. TAT activity was increased ($p \leq 0.01$ or 0.001) with respect to controls in the 2.5% Tyr group (↑123%) and in all mesotrione-treated groups (↑189-365%), although not dose-dependently. HPPD activity was decreased ($p \leq 0.05$ or 0.01) with respect to controls in the 2.5% Tyr group (↓54%) and in all mesotrione-treated groups (↓78-93%); the decrease was inversely proportional to the dietary tyrosine concentration. Relative liver weights were increased ($p \leq 0.05$ or 0.01) in all mesotrione-treated groups (↑10 -11%) and in the 0.5% Tyr group (↑6%), whereas absolute liver weights were increased ($p \leq 0.05$) only in the 100 ppm/0% Tyr (↑11%) and the 100 ppm/0.5% Tyr (↑8%). Relative kidney weights were increased in the 100 ppm/0.5% Tyr (↑8%) and 100 ppm/2.5% Tyr (↑15%) groups. At necropsy, cloudy eyes were observed in all mesotrione/tyrosine-treated animals (24/24 treated).

In conclusion, 100 ppm mesotrione in combination with tyrosine in the diet caused marked tyrosinemia and associated ocular lesions and changes in liver enzyme activities. In general, treatment with a combination of mesotrione and tyrosine caused more marked effects than treatment with either compound alone.

5) The objective of this study (MRID 44505113) was to assess the differences in systemic exposure from treatment with two different batches of mesotrione (ZA1296; Batch P8 [93.3% w/w] and Batch P11 [95.1% w/w]) administered for 7 days in the diet to male AlPk:APfSD rats (18/group) at levels of 1, 40, 25, 1250, or 5000 ppm. Exposure was compared from measurements of mesotrione in urine and blood plasma and from the dosing effect on plasma tyrosine concentrations. Total food consumption (for days 1 through 7) was also determined. Each parameter was determined on 3 rats/dose/batch.

The total amount of food consumed by each group was similar; there appeared to be no differences in palatability between the test diets. Systemic exposure comparisons based on

21

urinary excretion of mesotrione and its effect on plasma tyrosine concentrations showed no significant differences between Batch P8 and P11. There was a linear increase in area under the curve (AUC) and the maximum observed plasma concentrations (Cp_{max}) with increasing dose for both Batch P8 and Batch P11. A comparison of the AUC and the Cp_{max} of Batch P8 with Batch P11 suggested that at dose levels >1000 ppm, higher systemic exposure of Batch P11 may occur.
From the information provided, it was not clear whether or not the two batches of mesotrione were tested concurrently and whether or not the same animals were used to test the two batches of test material. The results of this special study are presented as an attachment to this overview (study report Figures 1 through 4, pages 8-11).

6) The objective of this study (MRID 44505116) was to investigate the tyrosinemia induced by mesotrione over a range of dose levels in male and female C57BL/10J$_f$AP/Alpk mice. Tyrosinemia occurs in animals fed mesotrione or other triketones due to an inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD). HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. HPPA is formed from tyrosine by the action of tyrosine aminotransferase (TAT). C57BL/10J$_f$AP/Alpk mice (20/sex/group) were fed diets containing mesotrione (96.8% a.i., Batch P17) at 0, 1, 10, 50, 100, 350, 1000, 3500, or 7000 ppm (equivalent to 0, 0.16/0.19, 1.69/1.94, 8.49/10.80, 17.95/20.46, 58.46/72.70, 179.27/214.88, 599.85/714.76, or 1222.53/1436.40 [M/F] mg/kg/day) for 90 days. Ten animals/sex/group were sacrificed at 1 and 4 weeks; the remainder (10/sex/group) were sacrificed at week 14.

Clinical signs, body weights, food consumption, plasma tyrosine levels, urine phenolic acid levels, kidney and liver weights, and liver TAT and HPPD levels were measured or recorded. Necropsies were performed at termination or upon the event of premature death.

Results of toxicological concern of this special study are presented as an attachment to this overview (Study Report Tables 6, 7, 8, 10, and 11, pages 64 through 68 and 82 through 85). There were no differences of toxicological concern in clinical signs, body weights, body weight gain (as calculated by the reviewers), and food consumption. Gross necropsy results were not reported. Differences in organ weights were minor and/or not time and dose-dependent and therefore not considered of toxicological concern.

Food utilization was decreased ($p \leq 0.05$ or 0.01) in 7000 ppm females for the overall treatment period (weeks 1-13, ↓22%), due primarily to a decrease at weeks 5-8 (↓41%). Free urinary phenolic acids were increased at week 13 in the 1-7000 ppm females (0.56-13.87 mg equivalent/mL in treated animals vs non detectable in controls) and the 10-7000 ppm males (4.20-15.52 mg equivalent/mL in treated animals vs 1.43-2.39 in controls). Conjugated phenolic acids were not detected in the male urine and detectable but not quantifiable due to very low levels in the female urine at ≥3500 ppm. Plasma tyrosine concentrations were elevated ($p \leq 0.01$) with respect to controls as follows: at week 1 in the 1-7000 ppm males (↑49-513%) and the 10-

22

7000 ppm females (↑110-451%); at week 4 in the 10-7000 ppm males (↑139-449%) and in the 1-7000 ppm females (↑40-493%); and at week 14 in the 10 and 100-7000 ppm males (↑87-257%) and the 10-7000 ppm females (↑69-316%).

TAT activity was increased ($p \leq 0.05$ or 0.01) with respect to controls as follows: at week 1 in the 100 and 1000 ppm males (↑29-33%) and the 50, 100, 1000, and 3500 ppm females (↑37-64%); at week 4 in the 350 ppm males (↑49%) and 1-7000 ppm females (↑47-104%); at week 14 in the 3500 ppm males (↑33%) and the 50-7000 ppm females (↑41-69%). HPPD activity was decreased ($p \leq 0.05$ or 0.01) with respect to controls as follows: at week 1 in all male (↓64-94%) and female (↓69-97%) treatment groups; at week 4 in all male (↓55-97%) and female (↓58-96%) treatment groups; at week 14 in all male (↓48-92%, excluding the 50 ppm males) and 100-7000 ppm female (↓60-75%) treatment groups.

In conclusion, treatment with increasing levels of mesotrione caused tyrosinemia with associated increases in urinary phenolic acids output and changes in liver enzyme activities.

7) The objective of this study (MRID 44505021) was to investigate the dose-response relationship for body weights and organ weight changes in male Alpk:AP$_f$SD rats. Male Alpk:AP$_f$SD rats (12/group) were fed diets containing mesotrione (95.1% a.i., Batch P11) at 0, 10, 20, 50, or 125 ppm (equivalent to 0, 0.9, 1.7, 4.3, or 10.7 mg/kg/day) for 90 days.

Clinical signs, body weights, food consumption, and kidney and liver weights were measured and/or recorded. Ophthalmoscopic exams were performed on all survivors prior to termination. Necropsies were performed at termination or upon the event of premature death.

Results of toxicological concern of this special study are presented as an attachment to this overview (Study Report Tables 6, 9, 10, and 11, pages 29 and 35 through 38). No treatment-related mortalities occurred. There were no differences of toxicological concern in body weights, body weight gain (as calculated by the reviewers), or food consumption. Gross necropsy results were not reported.

Opaque eyes were observed during clinical examinations in all treatment groups (3-9/12 each treated vs 0/12 controls). At ophthalmoscopic exam, the incidence of slight to marked hazy opacity or slight to marked opacity was increased in the all treatment groups (7/24, 12/24, 9/22, and 18/24 eyes examined in the 10, 20, 50, and 125 ppm groups, respectively, vs 1/24 controls). Corneal vascularization was also observed in all treatment groups (2/24, 8/24, 8/22, and 14/24 eyes examined in the 10, 20, 50, and 125 ppm groups, respectively, vs 0/24 controls). Ghost vascularization of the cornea was observed in the 10 (1/24 eyes examined vs 0/24 controls) and 50 (1/22) ppm animals. Food utilization was decreased in the 125 ppm group during weeks 9-13 (↓13%, $p \leq 0.05$).

Adjusted (to body) kidney weights were increased ($p \leq 0.01$) at termination in all treatment groups (↑8-10%), but not dose-dependently. Absolute kidney weights were increased only in the 50 ppm group (↑7%, $p \leq 0.05$). Adjusted liver weights were also increased ($p \leq 0.01$) non-dose-dependently in all treatment groups (↑12-14%). Absolute liver weights were increased ($p \leq 0.05$) in the 10 and 50 ppm groups only (↑9-11%).

In conclusion, treatment with mesotrione at 10, 20, 50, or 125 ppm caused dose-dependent changes in corneal opacity and corneal vascularization. Changes in adjusted (to body) kidney and liver weights occurred in all treatment groups, but these changes were not dose-dependent. There were no changes of toxicological concern in body weights.

8) In this special study, (MRID 44505029), mesotrione (100% purity and batch number Y06684/003/001) was administered for 28 days in the diet to CD rats and CD1 mice (5/group) at 1000, 3000 (mice only) 7000, or 16000 (rats only) ppm. The control animals (10/group) received control diet only. Standard cytochrome P450 (CYP) inducers were administered intraperitoneally (i.p.) for 4 days to Alpk:AP rats (3/group) and AP Swiss mice (3/group).

The objective of this study was to investigate the induction of hepatic CYP isoforms by mesotrione and compare the results with those of a series of standard CYP inducing agents including: β- naphthoflavone (BNF), phenobarbital (PB), dexamethasone (DEX), and methylclofenapate (MCP). The induction of CYP isoforms by the inducing agents was performed on Alpk:AP rats and AP Swiss mice because of animal supply problems. Food consumption, body weights, and clinical observations were performed throughout the study and at termination. Three CYP isoforms were quantified by immunoblotting. The use of monoclonal vs polyclonal antibodies for the immunoblotting assay was not specified. The specificities of the model substrates for three CYP isoforms were deduced from the effects of the standard inducers and their isoenzyme profiles after electrophoresis.

Food consumption, body weights, and clinical signs in both species and clinical chemistry parameters in the rat were not affected by treatment with mesotrione. In mice, treatment with mesotrione produced an increase ($p<0.05$) in plasma triglycerides at 3000 (↑41%) and 7000 ppm (↑45%), a decrease ($p<0.05$) in plasma alanine transaminase (ALT) at 7000 ppm (↓23%), and slight liver hypertrophy at 3000 (2/5 animals) and 7000 ppm (4/5 animals). In rats, treatment with mesotrione produced an increase ($p<0.05$) in relative (to body) liver weights in the 1000 (↑9%) and 16000 (↑11%) ppm groups and an increase in slight centrilobular hypertrophy at 16000 ppm (4/5 treated). Treatment with mesotrione had no effect on absolute or relative (to body) liver weights in mice or total CYP in either species. In mesotrione-treated rats, increases ($p<0.05$, 0.01, or 0.001) in activity of 4 CYP enzymes were observed at 7000 ppm (↑54-141%) and 16000 ppm (28-147%). In mice, increases ($p<0.01$) were observed in the activities of one CYP enzyme in the 3000 ppm (↑253%) and in two CYP enzymes in the 7000 ppm group (↑26-189%). In the 7000 and 16000 ppm rats, CYP isoform profiles showed minimal induction relative to controls in CYP 1A1 and its associated enzyme activities. In mice, minimal induction

was observed in CYP 2B1/2 (3000 and 7000 ppm groups) and CYP 3A1 (1000 and 3000 ppm groups) and their associated enzyme activity.

In rats, treatment with CYP inducers resulted in increased ($p<0.01$ or 0.001) absolute and relative liver weights. In mice, only treatment with MCP resulted in increased ($p<0.001$) absolute and relative (to body) liver weights; treatment with PB resulted in an increase ($p<0.05$) in relative liver weight. In general, the standard CYP inducers caused large increases in the activities of the CYP enzymes. The standard inducers produced moderate to marked increases in CYP 1A1, 2B1/2, and 3A1 isoform profiles of both species. BNF induced only 1A1 while PB and DEX induced both 2B1/2 and 3A1. Induction profiles for MCP were not analyzed. The results of this special study are presented as an attachment to this overview (study report Tables 1 through 9, pages 17-25).

In summary, treatment with mesotrione for 28 days resulted in slight increases in rat liver weights, slight microscopic liver hypertrophy in rats and mice, and minimal CYP induction in both species.

Metabolites

1) The objective of this study (MRID 44901706) was to investigate the toxicity of MNBA (97.1% a.i., Batch WRC 15483-30-1), a triketone analog, in male and female rats when administered by gavage for 28 consecutive days. Adult male and female Alpk:ApfSD rats (5/sex/group) were gavaged daily with MNBA in corn oil at 0, 15, 150, or 1000 (limit dose) mg/kg/day for 28 days. Body weights, food consumption, functional observational battery (FOB), hematology, and blood clinical chemistry parameters were measured. The adrenals, brain, epididymides, heart, kidneys, liver, spleen, testes, and thymus were weighed. Clinical observations, histopathological examinations, and necropsies were performed.

Results of toxicological concern in this special study are presented as an attachment to this overview (Study Report Tables 8, 11, 12, 13, and 14, pages 50, 53, 54, 58, 60, 69, and 70). Clinical observations, body weights, food consumption, blood clinical chemistry parameters, FOB, gross pathology, and histopathological observations were unaffected by treatment with the test substance.

There was no effect on motor activity in males. A dose-dependent increase in motor activity for the overall observation period (50 minutes) was observed in all female treatment groups (15 mg/kg - ↑23%, p= not significant; 150 mg/kg/day - ↑37%, $p\leq0.05$; and 1000 mg/kg -↑57%, $p\leq0.01$).

The following observations were made, but due to the lack of corroborating evidence were considered of equivocal toxicological concern: 1) eosinophilia ($p\leq0.05$ or 0.01) was observed in all female treatment groups (↑68, 100, and 92% in 15, 150, and 1000 mg/kg/day groups, respectively); 2) absolute and adjusted (to terminal body weight) spleen weights were decreased

(↓12%, p≤0.05) in 1000 mg/kg/day females; and 3) adjusted (to terminal body weight) testes weights were increased (↑13%, p≤0.05) in 1000 mg/kg/day males.

In conclusion, MNBA, when administered to adult rats by gavage at up to 1000 mg/kg/day for 28 days, caused an increase in motor activity in females.

2) The objective of this study (MRID 44901711) was to investigate the inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD) by AMBA (a mesotrione metabolite; 99% purity, Y09476/001/001) in isolated liver cytosol from untreated male Alpk:Ap$_f$SD rats. HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. The liver cytosol samples were incubated with (i) AMBA at concentrations of 0.02 or 20μM, (ii) mesotrione (96.8% purity, batch P17) and NTBC (92% purity) each at concentrations of 0.02, 0.2, or 20μM (positive controls), and (iii) sodium phosphate buffer only (negative control). To assay HPPD activity, oxygen consumption rates (μL O_2/min) for background, control (negative and positive), and treated liver cytosol samples were calculated.

The results of this special study are presented as an attachment to this overview (study report Tables 1and 2, page15). The addition of AMBA at 0.02 and 20 μM to isolated rat liver cytosol resulted in 0 and 18.7% inhibition of HPPD, respectively. The addition of mesotrione or NTBC at 0.02μM resulted in 70-78% inhibition of HPPD, while concentrations of 0.2 and 20μM resulted in 99-100% inhibition. The Sponsor stated that structure-activity relationships also indicated that the metabolite would be a weak inhibitor of HPPD.

In conclusion, AMBA is a weak inhibitor of HPPD *in vitro* and it appears it will not perturb tyrosine catabolism *in vivo*. A more definite conclusion cannot be reached without knowing the expected *in vivo* concentration.

3) The objective of this study (MRID 44901712) was to investigate the inhibition of p-hydroxyphenylpyruvate dioxygenase (HPPD) by MNBA (a mesotrione metabolite; 97% purity, Y08636/004/001) in isolated liver cytosol from untreated male Alpk:Ap$_f$SD rats. HPPD is involved in the catabolism of tyrosine by catalyzing the oxidative decarboxylation and rearrangement of 4-hydroxyphenylpyruvate (HPPA) to homogentisate. The liver cytosol samples were incubated with (i) MNBA at concentrations of 0.02 or 20μM, (ii) mesotrione (96.8% purity, batch P17) and NTBC (92% purity) each at concentrations of 0.02, 0.2, or 20μM (positive controls), and (iii) sodium phosphate buffer only (negative control). To assay HPPD activity, oxygen consumption rates (μL O_2/min) for background, control (negative and positive), and treated liver cytosol samples were calculated.

The results of this special study are presented as an attachment to this overview (study report Tables 1 and 2, page15). The addition of MNBA at 0.02 and 20 μM to isolated rat liver cytosol

resulted in 0 and 7.2% inhibition of HPPD, respectively. The addition of mesotrione or NTBC at 0.02μM resulted in 70-78% inhibition of HPPD, while concentrations of 0.2 and 20μM resulted in 99-100% inhibition. The Sponsor stated that structure activity relationships also indicated that the metabolite will be at most a weak inhibitor of HPPD.

In conclusion, MNBA is a very weak inhibitor of HPPD *in vitro* and it appears it will not perturb tyrosine catabolism *in vivo*. A more definite conclusion cannot be reached without knowing the expected *in vivo* concentration.

Special Studies - Human

1) In a special study (MRID 44505114), healthy male human subjects (6/dose) were given a single oral dose of mesotrione (99.7% a.i.; Batch # Y06684/219) at 0.1, 0.5, or 4 mg/kg. Pre- and post-dosing urinalysis, hematology, and clinical chemistry parameters were measured, and a pulmonary function test, electrocardiogram, and corneal exam were performed pre-study and post-study. Plasma tyrosine levels were determined pre- and post-dosing from -72 to +96 hours of dosing.

The stated purposes of this study were to 1) identify suitable urinary markers to allow non-invasive monitoring of worker exposure to mesotrione and 2) to define the dose response for increased plasma tyrosine to mesotrione dose.

No effects were observed on hematology, clinical chemistry, or urinalysis parameters. No adverse effects were observed upon physical examination.

Dosing with mesotrione at 0.1, 0.5, or 4 mg/kg elevated plasma tyrosine starting at 2 hours post-dosing and remaining elevated until between 12 and 24 hours (0.1 and 0.5 mg/kg) or between 36 and 48 hours (4 mg/kg) post-dosing. The mean plasma tyrosine levels peaked at 5 hours post-dosing in the 0.1 mg/kg group (129 nmol/mL), at 6 hours post-dosing in the 0.5 group (152 nmol/mL), and at 8 hours post-dosing in the 4 mg/kg group (289 nmol/mL).

At 0.1 mg/kg, plasma tyrosine maximums ranged from 91.1 to 161 nmol/mL during the 24 hour period following dosing vs. 68.6 to 106 nmol/mL during pre-dosing.

At 0.5 mg/kg, plasma tyrosine maximums ranged from 121 to 210 nmol/mL during the 24 hour period following dosing vs. 65.1 to 102 nmol/mL during pre-dosing.

At 4 mg/kg, plasma tyrosine maximums ranged from 241 to 420 nmol/mL during the 24 hour period following dosing vs. 95.3 to 127 nmol/mL during pre-dosing.

Mean AUC values were increased in a dose-dependent manner during the 24 hours immediately following dosing (day 0) with respect to the 24 hour period immediately preceding dosing (day -1): 0.1 mg/kg - (↑42%); 0.5 mg/kg - (↑84%); and 4 mg/kg - (↑130%) mg/kg subjects. For the period from 24 to 48 hours following dosing (day 1), plasma tyrosine AUC values remained slightly elevated with respect to day -1 levels (↑16%, ↑32%, and ↑27% in 0.1, 0.5, and 4 mg/kg

subjects, respectively). For the periods from 48 to 72 hours (day 2) and 72 to 96 hours (day 3) following dosing, plasma tyrosine AUC values were only slightly elevated in 0.1 (↑5-7%) and 4 mg/kg (↑5-8%) treated subjects, whereas AUC values remained elevated in 0.5 mg/kg subjects (↑20-22%).

This study is classified **unacceptable (non-guideline)** and does not satisfy the purposes for which it was intended. No data were presented regarding identifying suitable urinary markers to allow non-invasive monitoring of worker exposure to mesotrione and the dose response for increased plasma tyrosine to mesotrione dose was not analyzed statistically.

2) In a special study (MRID 44505115), 10 healthy human male subjects were given a single oral dose of NTBC (purity not reported) at 1 mg/kg in either liquid or capsule (Period 1). After approximately 14 days, patients who received the liquid dose were dosed by capsule and vice versa (Period 2). Plasma tyrosine concentrations were determined from 0–120 hours post-dosing for both periods.

The purpose of the study was to compare the bioavailability of NTBC at therapeutic levels in two different formulations. This report constitutes the analytical portion of the study. The clinical portion of the study was performed at Cardiff Clinical Trials Ltd, Cardiff Medicentre, Heath Park, Cardiff, UK.

A single dose of NTBC increased mean plasma tyrosine levels in a linear fashion until approximately 48 hours after dosing, at which time tyrosine levels plateaued. At maximum levels, plasma tyrosine was 1039.6 nmol/mL. This level was increased 935% from a mean pretreatment level of 100.4 nmol/mL. After 14 days recovery, mean plasma tyrosine concentrations had not returned to pre-dosing levels and were still high (808.4 nmol/mL). Administration of the second dose of NTBC increased plasma tyrosine levels to a mean maximum level of 1050.3 nmol/mL. This level is approximately equal to the maximum reached after the first dose, and is 946% greater than the first pre-dose measurement.

Dosing formulation appeared to not have an effect on the bioavailability of NTBC.

This is study is classified **acceptable (non-guideline)** and does satisfy the purposes for which it was intended.

3) In this special study (MRID 44920803), ZA 1296 (mesotrione (ZA1296; Lot/Batch # WF2515, 9.1% a.i. and WF2381, 39.8% a.i.) was administered as a single dermal application to 18 volunteer human males. The volunteers were assigned to one of 3 study phases (6 volunteers/phase) and received nominal doses of 4mg (5ug/cm^2 - WF2515), 4mg (5ug/cm^2 - WF2381), or 25.6mg (32ug/cm^2 - WF2381) mesotrione. The application sites were washed after 10 hours. The volunteers were monitored for 5 days post-dosing, and then participated in post-study medical examinations 4-7 days after discharge. The objective of this study was to determine the urine and plasma concentrations of mesotrione following dermal application, and

to define the dose-response relationship for increased plasma tyrosine. The study was conducted in accordance with the Declaration of Helsinki (1964) including all amendments up to and including the South Africa revision (1996).

No treatment-related changes in vital signs, ophthalmoscopic observations, or hematology, clinical chemistry, or urinalysis parameters were observed. No quantifiable concentrations of mesotrione were detected in the plasma or in urine samples from phases 1 and 2. Detectable levels occurred in urine at 2-5/13 timepoints in phase 3 and ranged from 5-9 ng/mL vs. a detection limit of 5-6 ng/mL. Comparison of tyrosine post-dose area under the curve (AUC) and Cp_{max} values with pre-test values indicated that dermal application of mesotrione had no discernable effect on plasma tyrosine concentration.

The number of volunteers reporting mild, transient itching was increased in phase 3 (4 volunteers at 3 timepoints) compared to phases 1 and 2 (1 volunteer at 1 timepoint, each). Other symptoms included reports of a burning sensation 30 minutes after dose application (1 volunteer in phase 1) and a stinging sensation 9 hours after dose application (1 volunteer in phase 2); these symptoms were also mild and transient in nature. All volunteers in phase 1 (14 timepoints), 1 volunteer in phase 2 (2 timepoints), and 6 volunteers in phase 3 (10 timepoints) experienced erythema.

Results of the tape strip analyses indicated that the percentage of the applied dose remaining at the application site decreased between the 10-hour (35-54.7% remaining) and 24-hour (10.3-20.0% remaining) timepoints in all phases of the study. Analysis of the swabs from each phase of the study indicated that a large proportion of the applied dose was removed from the application site during the washing procedure (36.8-49.6%). T-shirt analysis demonstrated that additional test-substance was transferred from the application site to the t-shirts between 10 and 24 hours post-dosing for each phase (8.65-29.7%). The tape strip results could not be analyzed as the majority were below the limit of detection.

Mesotrione appeared to remain in the stratum corneum and was easily removed by washing or contact with clothing.

The submitted study is classified as **acceptable/non-guideline**.

4.3 Prenatal Developmental Toxicity

Adequacy of data base for Prenatal Developmental Toxicity: The data base for prenatal developmental toxicity is considered complete and no additional studies are required at this time. Oral rat, mouse, and rabbit developmental studies showed an increased susceptibility of the fetus to mesotrione *in utero*. Delayed ossification was seen in the fetuses at doses below those at which maternal toxic effects were noted. Maternal toxic effects were decreased body weight gain and food consumption in the rat and abortions and GI effects in the rabbit. No maternal toxic effects were noted in the mouse. In a special range-finding study, both litter size and pup

viability were decreased following treatment of the maternal animals with 2500 ppm mesotrione co-administered with 0.5% and 1% tyrosine; males appeared to be more susceptible to the effects of treatment.

870.3700a Prenatal Developmental Toxicity Study - Rodent

1) In a developmental toxicity study (MRID 44920801), mesotrione (96.8% a.i., Lot#: P17) in deionized water was administered to pregnant Alpk:AP$_f$SD rats (24/dose) at dose levels of 0, 100, 300, or 1000 (limit dose) mg/kg/day by gavage on gestation days (GDs) 7 through 16. All dams were sacrificed on GD 22. No premature deaths occurred during the study.

Treatment-related toxicity was characterized by reduced body weight gains and food consumption at the mid- and high-dose levels. When compared to concurrent controls, no treatment-related changes in absolute body weight, gravid uterine weight, Cesarean section parameters, or gross pathology were noted at any dose level tested. Urine staining and colored feces were not considered adverse clinical effects.

At 1000 mg/kg, pink and/or purple colored feces (130 incidents in 22/24 animals vs 0/24 controls) and dry or wet staining with urine (29-39 observations in 6-8/24 animals vs 0/24 controls) were observed. Body weight gains, as calculated by reviewers, were reduced during the overall treatment interval (↓20%, GDs 8-16, not analyzed for statistical significance). In addition, decreases were noted in food consumption during GDs 7-16 (↓14-18%, $p \leq 0.01$). Body weights and food consumption increased in all dose groups including controls during the post-treatment interval.

At 300 mg/kg, an increased incidence of pink and/or purple colored feces was noted (35 observations in 10/24 animals vs 0/24 controls). Body weight gains were reduced during the overall treatment interval (↓17%, GDs 8-16). Decreases were noted in food consumption during GDs 7 through 16 (↓8-13%, $p \leq 0.01$).

At 100 mg/kg, no treatment-related adverse effects were noted.

The maternal LOAEL is 300 mg/kg/day, based upon decreased body weight gains during treatment and decreased food consumption. The maternal NOAEL is 100 mg/kg/day.

Developmental toxicity was characterized by decreased mean fetal weight (↓6%, $p \leq 0.01$) at the 1000 mg/kg/day level and a dose-dependent increase in incidences of decreased ossification of vertebral centra and of the *manus* and *pes* at all dose levels. Incidences of "non-ossification" of several cervical centra ranged from 2 - 87% (25 - 100%)[% fetal incidence (% litter incidence)] as compared to controls [0.2 - 22% (4.2 - 88%)]. Reduced ossification was seen in other skeletal structures such as cervical vertebra arches, vertebra transverse processes, otontoids, and calcaneum. When compared to mean scores for the controls, increased mean *manus* and *pes* scores/litter for all treatment groups were observed (↑4-11%). It should be noted that this shift toward reduced ossification was also observed in mice administered the test substance (MRID 44920802).

The developmental LOAEL is 100 mg/kg/day, based on delays in skeletal ossification and changes in *manus/pes* ossification assessments. The developmental NOAEL was not established.

Even though a developmental NOAEL was not established and no dose rationale was provided. this developmental toxicity study is classified **acceptable/guideline (§83-3[a])**, and satisfies the guideline requirements for a developmental toxicity study in the rat as per the Hazard Identification Assessment Review Committee (March 13, 2001).

2) In a developmental toxicity study (MRID 44920802 and 44901708), mesotrione (96.8% a.i., Lot #: P17) in water was administered to pregnant Alpk:AP$_f$CD-1 mice (26/dose) at dose levels of 0, 10, 60, 150, or 600 mg/kg/day by gavage on gestation days (GDs) 5 through 18. All dams were sacrificed on GD 19. One 60 mg/kg female was sacrificed *in extremis* on GD 12 following the observation of a subcutaneous mass on the left anterior thorax on GD 9 and subsequent reduced body weight and food consumption. No other premature deaths occurred during the study.

When compared to concurrent controls, no treatment-related clinical signs, changes in body weight or adjusted body weights (using GD 5 body weight as a covariant), gravid uterine weight, food consumption, gross pathology, or reproductive parameters were noted at any dose level tested.

The maternal LOAEL was not observed.
The maternal NOAEL was ≥ 600 mg/kg/day.

At 600 mg/kg, a treatment-related pattern toward decreased ossification of the cervical vertebrae centra was observed. In addition, a number of other delays in ossification or variations were apparent at the high dose level (see Table 4c).

The developmental LOAEL is 600 mg/kg/day based on the pattern toward decreased ossification of the cervical vertebrae centra.
The developmental NOAEL is 150 mg/kg/day.

This developmental toxicity study is classified **acceptable (§83-3[b])** and does satisfy the guideline requirement for a developmental toxicity study in the mouse.

870.3700b Prenatal Developmental Toxicity Study - Rabbit

In a developmental toxicity study with rangefinding (MRIDs 44901707 and 44505032), mesotrione (96.8% a.i., Lot #: P17) in deionized water was administered to pregnant New Zealand White rabbits (20/dose) at dose levels of 0, 100, 250, or 500 mg/kg/day by gavage on gestation days (GDs) 8 through 20. All does were sacrificed on GD 30.

A single 100 mg/kg animal was found dead on GD 4 and abnormal GI tract contents were observed at necropsy. One 250 mg/kg animal was sacrificed *in extremis* on GD 22 after displaying clinical signs including diarrhea, subdued behavior, thin appearance, and severe weight loss. Five does were sacrificed after showing signs of abortion as follows: one 100 mg/kg on GD 29; two 250 mg/kg on GD 25 or 28; and two 500 mg/kg on GD 23 or 25.

At 500 mg/kg/day, observations included the following: blood on the tray (6 observations in 4/20 animals vs 1 incidence in 1/20 controls); few feces on the tray (25 incidences in 7/20 animals vs 3 observations in 2/20 controls); no feces on the tray (6 observations in 3/20 animals vs 0/20 controls); and red to brown colored urine (41 observations in 12/20 animals vs 1 incident in 1/20 controls); decreases (NS) in food consumption during GDs 14-20 (↓12-24%) and increases during post-treatment (↑39%, GDs 26-30, $p \leq 0.01$); a decrease (NS) in gravid uterine weight (↓15%); and a decrease (NS) in the number of implantations/doe (↓13%). Food consumption was also reduced at both the 500 and 250 mg/kg/day dose levels with rebounds observed during the post dosing period.

At 250 mg/kg/day, findings observed included the following: few feces on the tray (11 observations in 4/20 animals vs 3 observations in 2/20 controls); no feces on the tray (4 observations in 3/20 animals vs 0/20 controls); red to brown colored urine (26 incidents in 12/20 animals vs 1 incident in 1/20 controls); and decreases (NS) in food consumption during GDs 14-20 (↓14-15%) and increases (NS) during the post-treatment interval (↑12%, GDs 26-30).

At 100 mg/kg/day, findings observed that were considered to be possibly treatment-related included few feces on the tray (6 observations in 5/20 animals vs 3 observations in 2/20 controls) and red to brown colored urine (14 incidents in 6/20 animals vs 1 incident in 1/20 controls).

When compared to concurrent controls, no treatment-related changes in body weight, adjusted body weight, or gross pathology were noted at any dose level tested.

NOTE: Due to the increase in preimplantation loss noted in the mid and high dose levels, these groups may have received test material prior to the completion of implantation. This would be a confounding factor which may interfere with the proper assessment of maternal toxicity in this study.

The maternal LOAEL is 250 mg/kg/day, based on abortions and clinical signs of toxicity. The maternal NOAEL is 100 mg/kg/day.

At 100, 250, and 500 mg/kg/day, skeletal examination (reported as [% fetal incidence (% litter incidence)]) revealed a shift toward a decreased degree of ossification of the 7th cervical vertebra transverse process, when compared to concurrent controls, as evidenced by i) a decreased ($p \leq 0.05$ or 0.01, fetal incidences only) incidence of a "partially ossified" transverse process at the low- [0.8 (7.1)], mid- [0.7 (5.9)], and high-dose levels [0 (0)] vs controls [6.7 (22.2)] and ii) a decreased (NS) incidence of a "fully ossified" transverse process in the 7th cervical vertebra in all

treated groups [0 (0)] vs controls [3.3 (11.1)]. In addition, statistically significant decreases in the ossification (fetal incidence only) of the odontoid was observed in all dose groups as compared to controls. Conversely, an unexpected trend toward more complete ossification of the 5th sternebra was observed and was demonstrated by i) a dose-dependent decrease ($p \leq 0.01$, fetal incidences only) in the number of animals exhibiting a "partially ossified" 5th sternebra at the low- [32.3 (92.9)], mid- [28.9 (76.5)], and high-dose levels [24.6 (62.5)] vs controls [52.0 (94.4)] and ii) a decrease ($p \leq 0.05$ or 0.01, fetal incidences only) in the number of animals exhibiting a "nonossified" 5th sternebra at the low- [3.2 (14.3)], mid- [3.4 (11.8)], and high-dose levels [4.2 (25.0)] vs controls [12.7 (33.3)]. In addition, statistically significant ($p \leq 0.01$) increases in the fetal incidence of 13 full ribs and 27 pre-sacral vertebra were noted at all dose levels.

An increase in preimplantation loss ($p \leq 0.01$) was noted at the high-dose level in the submitted report, but errors were observed in the calculation, and therefore, the percent losses were recalculated by reviewers. As a result of the Agency contractor re-calculation, increases were observed at both the mid- (↑42%) and high-dose (↑152%) levels. The observation of preimplantion loss in several dose groups would suggest dosing began in these groups prior to the completion of the implantation process. This introduces a confounding factor into study interpretation.

At 500 mg/kg, a decrease (NS) in the number of live fetuses/doe (↓11%) was observed. This may be associated with the preimplantation loss noted in this dose group.

There were no treatment-related external or visceral effects noted at any dose level. Treatment-related changes observed in the *manus* ossification data included statistically significant increases in the proportion of fetuses with scores of 5 at the mid and high dose levels.

The developmental LOAEL is 100 mg/kg/day, based on delayed ossification of the 7th cervical transverse process and odontoid and increases in extra full 13th ribs and 27 pre-sacral vertebra. Hence, a developmental NOAEL was not established.

Since a developmental NOAEL was not established and because dosing was apparently initiated in several dose groups prior to the completion of implantation (resulting in increased preimplantation loss), this developmental toxicity study is classified **unacceptable/ not upgradeable** (§83-3[b]) and does not satisfy the guideline requirement for a developmental toxicity study in the rabbit.

Range-finding study

The stated objective of this study (MRID 44505112) was to investigate the effects of mesotrione (96.8% purity, batch P17) in conjunction with dietary tyrosine on litter size and pup viability in Alpk:Ap$_f$SD rats. One control group and 7 test groups (20 time-mated females/group) received mesotrione and/or L-tyrosine in the diet as follows: Group 1 was the control group, Group 2

received 0 ppm mesotrione + 0.5% tyrosine, Group 3 received 0 ppm mesotrione + 1% tyrosine, Group 4 received 0 ppm mesotrione + 2% tyrosine, Group 5 received 2500 ppm mesotrione only, Group 6 received 2500 ppm mesotrione + 0.5% tyrosine, Group 7 received 2500 ppm mesotrione + 1% tyrosine, and Group 8 received 2500 ppm mesotrione + 2% tyrosine. Diets were administered continuously beginning on gestation day (GD) 0. Clinical signs, body weights, food consumption, and plasma tyrosine levels were measured in the dams; offspring were counted, examined for clinical signs, and weighed. Any animal which failed to litter by GD 25 or experienced difficulties with parturition was sacrificed and discarded.

The results of this special study are presented as an attachment to this overview (Study Report Tables 1 through 5, pages 9 through 15). There were no differences of toxicological concern in pup body weights.

At 48 hours after the start of treatment, tyrosine levels in groups 2-8 were increased (↑9-1807%, not analyzed for statistical significance). During clinical observation, the following findings were noted: eye opacity in groups 6 (7/20 treated), 7 (15/20 treated), and 8 (8/20 treated); hunched posture in 5/20 group 8 animals; and piloerection in groups 5 (4/20 treated), 6 (12/20 treated), 7 (16/20 treated), and 8 (14/20 treated). All group 8 females displayed severe corneal lesion which necessitated the termination of the entire group on GDs 8-11. Decreased ($p<0.001$) body weights were observed in group 8 animals on GDs 4 and 7 (↓11-13%).

Increased incidences of whole litter losses between PNDs 1-5 occurred in groups 6 and 7 (4/17 and 8/18, respectively). Decreased litter size on PND 1 was noted in group 7 (↓16%, not statistically significant [NS]). The mean number of pups found dead on PND 1 was increased in group 7 (↑435%, $p<0.05$). In groups 6 and 7, a decreased number of live male pups (mean number live pups litter = 5.35 and 4.83, respectively, vs 7.05 controls, $p<0.05$) and an increased number of dead male pups (mean number dead/litter = 0.35 and 0.67, respectively, vs 0 controls, $p<0.05$) were noted on PND 1. In groups 6 and 7, the following findings ($p<0.05$ or 0.001) were noted on PND 5: decreased number of live pups/litter (↓23 and 53%, respectively); increased percent deaths (↑225 and 525%, respectively); decreased number of male pups/litter (↓44 and 61%, respectively); increased percent deaths in males (↑372 and 946%, respectively); decreased number of female pups/litter (group 7 only, ↓41%); and increased percent deaths in females (group 7 only, ↑192%). An additional finding in group 6 included an increase in percent deaths in the females (↑80%, NS). In group 5 on PND 5 the following findings (NS) were noted: decreased number of live pups/litter (↓13%); increased percent deaths (↑110%); increased percent deaths in the males (↑137%) and in the females (↑111%).

In conclusion, both litter size and pup viability were decreased following treatment of the maternal animals with 2500 ppm mesotrione co-administered with 0.5% and 1% tyrosine; males appeared to be more susceptible to the effects of treatment.

4.4 Reproductive Toxicity

Adequacy of data base for Reproductive Toxicity: The data base for reproductive toxicity is considered complete and no additional studies are required at this time. Multi-generation reproduction studies also showed an increased susceptibility of the young to mesotrione. In the mouse, the young exhibited significant tyrosinemia and ocular discharge at doses below those at which parental toxic effects were noted. Additionally, cataractous changes were observed histologically at the high dose level (>1000 mg/kg). At the LOAEL for parental effects, tyrosinemia and increased kidney weights were observed; ocular lesions in both sexes and decreased body weights in females were observed at higher dose levels. In addition, one F_1 male and one F_1 female had retinal detachment with marked cataractous changes at the highest dose tested (>1000 mg/kg). In the rat, no NOAEL was determined for parental effects (tyrosinemia and increased liver weights) or offspring systemic effects (tyrosinemia); however, the tyrosinemia was much more severe in the young than in the adults. Decreased litter size was noted at the next highest dose. Additionally in the adult rats and offspring, macroscopic and microscopic ocular opacity/cloudiness, increased kidney weights and an increased incidence of hydronephrosis were also observed at >LOAEL.

870.3800 Reproduction and Fertility Effects - Rodent

1) In a 3-generation reproduction study (MRID 44505033), mesotrione (96.8% a.i., lot # P17) was administered in the diet continuously to 3 generations of Alpk:AP$_f$SD (Sprague-Dawley) rats (26 rats/sex/dose) at dose levels of 0, 2.5, 10, 100, or 2500 ppm (equivalent to 0, 0.3/0.3, 1.1/1.2, 11.7/12.4, or 287.7/311.4 mg/kg/day [M/F] in the P and F_1 animals). The P and F_1 animals were exposed to the test substance for approximately 10 weeks prior to mating. At approximately 14 weeks after selection, the F_2 animals were subdivided into a continuous treatment group (12 animals/sex/dose) and a recovery group (14 animals/sex/dose), which received control ration only. At approximately 4 weeks after subdivision, these groups were mated to produce the F_3 generation. Excluding the recovery group, exposure of all animals to the test material was continuous throughout the study.

There was no evidence of treatment-related changes in mortality, body weight gains, food efficiency, or reproductive performance observed in the P or F_1 adults at any dose. Decreased food consumption during lactation, increased incidences of ocular opacity, cloudiness, keratitis, and increased corneal vascularization, and increased bilateral hydronephrosis were observed in the100 and 2500 ppm groups. The severity of these effects was greater than the 10 ppm groups in a dose-dependent manner. At 10 ppm, differences in food consumption were observed in F_1 dams (↓15-17%, LWs 3 and 4).

Ophthalmologic findings were observed during clinical observation, at necropsy, and at histological examination in the treatment groups, but not in controls, except as noted. During clinical observation, cloudy/opaque eyes were observed in the following groups: F_1 males (1/26); F_2 males before subdivision (1/26); and F_2 males in the recovery group (1/14).

At necropsy, opaque/cloudy eyes were observed in the F_1 males (1/26 treated) and F_2 recovery males (1/14 treated). At histological examination, keratitis was observed in F_1 males (5/26 treated). Corneal vascularization was observed in F_1 males (5/26 treated). Bilateral hydronephrosis at histological exam and under 2X magnification was observed in F_1 males (10/26) and F_2 continuous treatment males (2/12). No incidences were noted in the controls for any of these groups.

Increases in absolute and adjusted (to body weight) liver weights were observed. Liver weights were increased in the P, F_1, and F_2 groups as follows: In the 10, 100, and 2500 ppm P males and 100 and 2500 ppm P females, in the 10, 100, and 2500 ppm males, in the 2.5, 10, 100, and 2500 ppm F_2 continuous treatment males, and in the 2500 ppm F_2 recovery group females. Absolute and/or adjusted (to body weight) kidney weights were increased in all 2500, 100, and 10 ppm male groups, including the recovery groups. Increased kidney weights were only observed in 2500 ppm P and recovery females. The incidence of bilateral hydronephrosis at terminal necropsy as determined under 2X magnification was increased in 2500 and 100 ppm males and females and F_2 continuous treatment males and in the 10 ppm males. At histological examination, the incidence of minimal to marked bilateral hydronephrosis was increased in 2500 ppm F_1, F_2 continuous treatment, and F_2 recovery males and females; in 100 ppm F_1 males and females, F_2 continuous treatment and recovery males; and in 10 ppm F_1 males. The nephrotoxicity was apparently not reversed in the recovery groups.

Plasma tyrosine levels were significantly increased in F_2 adult males under continuous treatment at all treatment doses during the pre-mating interval and at termination (↑569 - 2478%). Levels were significantly increased in F_2 adult females under continuous treatment at 10 ppm and above during the pre-mating interval and at termination (↑289 - 285%). Animals of both sexes in the recovery groups had similar plasma tyrosine levels as the controls at all doses.

The LOAEL for systemic parental toxicity is 2.5 ppm (equivalent to 0.3 mg/kg/day for both sexes) based on significantly increased plasma tyrosine levels and increased liver weights in F_2 males. No NOAEL was determined.

There was no evidence of treatment-related changes in body weights or body weight gains in the F_1 or F_2 litters at any dose.

A pattern of ocular toxicity consisting of macroscopic and microscopic ocular opacity/cloudiness was observed in all offspring generations. No opaque/cloudy eyes were observed in control animals at any time. An increase in the number of pups with cloudy eyes was observed in all 2500 groups and in the 100 ppm F_2 group (% pups[% litters]) with a range of 26-72 (36-61). An increase in the number of pups with ocular discharge was observed in the 2500 ppm F_1 (11[26] treated vs 0.4[4] controls) and F_2 litters (8[15] treated vs 0.5[5] controls), and 100 ppm F_2 litters (4[16]) but the effect was not observed in the F_3 litters.

At gross necropsy, an increase in the incidence of opaque or cloudy eyes was observed in the 2500 ppm F_1 and F_2 males and females and 100 ppm F_2 males. Closed eyelids were also observed in 2500 ppm F_1 males (18/54 treated vs 0/44 controls) and females (7/42 treated vs 1/49 controls). An increase in the incidence of minimal to marked ocular keratitis was observed at histological examination in the 2500 ppm P and F_1 males and females (18-26/26 treated vs 0/26 controls). Keratitis was also observed in 100 ppm P males and females (8-11/26 treated) and F_1 males and females (23-26/26 treated). Increased minimal to moderate corneal vascularization was observed in 2500 ppm P and F_1 males and females (16-26/26 treated vs 0/26 controls). Corneal vascularization was also observed in 100 ppm P males and females (7-8/26 treated). F_1 males and females (12-26/26 treated).

A pattern of nephrotoxicity consisting of increased kidney weights and increased macroscopic and microscopic renal hydronephrosis was observed in the pups. There was an increase in the relative kidney weights in F_2 males (↑11%) and relative and absolute kidney weights (↑15% each) in F_2 females. At gross necropsy, the incidence of bilateral renal pelvic dilatation was increased in the 100 and 2500 ppm F_3 continuous treatment males (15-18% in treated vs. 0% in controls). At histological examination, minimal to marked bilateral hydronephrosis was increased in the 100 and 2500 ppm F_1 and F_2 males and females (8-15% treated vs 1-4% controls), in the 10 ppm F_1 and F_2 males and females (5-7% treated) and in the 100 and 2500 ppm F_3 continuous treatment males and females (12-33% treated vs 2-4% controls). In the recovery animals, the incidences of bilateral hydronephrosis were low and similar to controls. The incidence of bilateral hydronephrosis at terminal necropsy, as determined under 2X magnification, was increased as follows: 100 and 2500 ppm F_1 , F_2, and F_3 continuous treatment males and females; 10 ppm F_1 males and females; and 10 ppm F_2 males.

Plasma tyrosine levels were significantly increased in F_3 male pups under continuous treatment at all treatment doses (↑979 - 2374%). Levels were significantly increased in F_3 female pups under continuous treatment at 100 and 2500 ppm (↑633 - 960%). Animals of both sexes in the recovery groups had similar plasma tyrosine levels as the controls at all doses.

The LOAEL for systemic offspring toxicity is 2.5 ppm (equivalent to 0.3 mg/kg/day for both sexes) based on significantly increased plasma tyrosine levels in F_3 male pups. No NOAEL was determined.

Mean litter size was decreased 20-45% compared to the controls throughout lactation in all 2500 ppm groups, including F_3 recovery litters. Mean litter size was also decreased in the 10 ppm (↓19-23%, PND 8-29) and 100 ppm (↓20-22%, PND 5-29) F_2 litters. At 2500 ppm, the livebirth index was decreased in the F_2 (↓6%) and F_3 continuous treatment (↓12%) litters and the day 22 viability index was decreased in the F_1 and F_2 litters (↓16% each). The proportion of litters with whole litter losses was increased in the 2500 ppm F_2 litters (7/20 treated vs 1/21 controls). There was no significant difference in the F_1 and F_3 continuous treatment litters. Whole litter weights were decreased throughout lactation in the 2500 ppm F_1, F_2, and F_3 continuous treatment litters (↓19-49%, PND 1-29); at PND 1 in 2500 ppm F_3 recovery litters (↓29%); beginning at

approximately the first week of lactation in the 100 ppm F_1 and F_2 litters (↓13-21); and at PND 11, 15, and 29 in the 10 ppm F_2 litters (↓18%).
The LOAEL for reproductive toxicity is 10 ppm (equivalent to 1.1/1.2 mg/kg/day [M/F]) based on decreased F_2 mean litter size. The reproductive NOAEL is 2.5 ppm (equivalent to 0.3 mg/kg/day for both sexes).

Even though no systemic NOAEL was determined for parents or offspring, the reproductive study is determined to be **acceptable/guideline (§83-4[a])** and satisfies the guideline requirement for a multigenerational reproductive toxicity study in rats as per the Hazard Identification Assessment Review Committee (March 13, 2001).

2) In a 2-generation reproduction study (MRID 44505034), mesotrione (96.8% a.i., lot # P17) was administered in the diet continuously to Alpk:APfCD-1 mice (26 mice/sex/dose) at dose levels of 0, 10, 50, 350, 1500, or 7000 ppm (equivalent to 0, 2.1/2.4, 10.1/11.7, 71.4/82.5, 306.7/362.7, or 1455.5/1652.3 mg/kg/day [M/F] in the P and F_1 animals). The P animals were exposed to the test substance beginning at approximately 3 weeks of age and exposure lasted for approximately 8 weeks prior to mating. F_1 pups selected (26/sex/dose) to produce the F_2 generation were exposed to the same dosage as their parents beginning on postnatal day (PND) 29 and continuously throughout the rest of the study. F_1 animals were administered the test article for approximately 8 weeks prior to mating to produce the F_2 animals. Mating to produce an F_{2b} generation was not performed. Exposure of all animals to the test material was continuous throughout the study. The analytical data indicated that the mixing procedure was adequate and the variation between nominal and actual dosage to the study animals was acceptable.

There were no statistically significant and treatment-related changes in mortality, or reproductive performance observed in the P or F_1 adults. Adjusted (to LD 1) body weights were decreased during lactation in 7000 ppm P dams on LDs 5, 8, and 15 (↓3-13%). Body weights were decreased (↓9-12%) in 7000 ppm F_1 dams on LDs 1 (absolute) and 15 (adjusted). Food consumption was decreased in the 7000 ppm P dams throughout lactation (↓15-28%) and in 7000 ppm F_1 dams during weeks 2, 3, and 4 of lactation (↓10-33%).

An increase in the incidence of opaque eyes in 7000 ppm F_1 males (4/26 treated vs 0/26 controls) and females (6/26 treated vs 0/26 controls) was observed. Opaque eyes were also observed in a single animal from each of the following groups: 7000 ppm P males, 10 and 350 ppm F_1 males, and 1500 ppm F_1 females. At necropsy, an associated increase was observed in the incidence of grossly visible opaque/cloudy eyes (4 - 20% treated vs. 0 - 4% controls) in 7000 ppm P males, P females, F_1 males and F_1 females. In addition, upon histological examination, an increase was observed in the incidence of minimal to marked unilateral and bilateral ocular cataractous change in the 7000 ppm P males (unilateral - 3/26 treated vs 0/26 controls), F_1 males (unilateral - 7/25 treated, bilateral 1/25 treated vs 0/26 controls), and F_1 females (unilateral - 4/26 treated, bilateral 1/26 treated vs 0/26 controls). Retinal detachment with marked cataractous change was also observed in one of the 7000 ppm F_1 males and females; whether this lesion was a primary effect of mesotrione on the eye is unclear.

Absolute kidney weights were statistically significantly increased in F_1 males at 350 ppm and above. Percent increases above controls were 11, 8, and 13% for 350, 1500, and 7000 ppm, respectively. Relative kidney weights were also statistically significantly increased at 350 ppm and above in males. Percent increases above controls were 6, 8 and 17% for 350, 1500, and 7000 ppm, respectively. In females, relative kidney weights were statistically significantly increased at 50 ppm and above, but percent increases on ranged from 4 to 9% above controls.

The LOAEL for parental toxicity is 350 ppm (equivalent to 71.4/82.5 mg/kg/day [M/F]) based upon dose-related and statistically significant increases in kidney weights in F_1 males. The increased kidney weights may be associated with tyrosyluria since tyrosinemia was observed at all dose levels in F_1 animals (exposed in utero) in this study. Ocular lesions in both sexes and decreased body weights in females were observed at higher dose levels. The NOAEL for parental toxicity is 50 ppm (equivalent to 10.1/11.7 mg/kg/day [M/F]).

No treatment-related effects on mortality or viability were observed at any time in the F_1 and F_2 litters. An increase in the number of pups with opaque eyes (6 pups in 1 litter vs 0 controls) and pups with eye(s) shut (7 pups in 4 litters vs 0 controls) was observed in the 7000 ppm F_2 pups. An increase in the number of pups with ocular discharge was observed in the 7000 ppm dose groups (F_1 and F_2 - 11 pups in 5 litters each vs 0 controls). A slight increase in the number of pups with ocular discharge was also observed in the other F_1 (1-5 pups/group in 1-3 litters/group in the 10, 50, 350, and 1500 ppm groups) and F_2 litters (2-3 pups/group in 2-3 litters in the 10, 350, and 1500 ppm groups). At necropsy, an increase was observed in the incidence of opaque or cloudy eyes in 7000 ppm (10/33 treated vs 0/30 controls) and 1500 ppm (4/30 treated) F_2 males. Upon histological examination, an associated increase was observed in the incidence of microscopic minimal to marked unilateral and/or bilateral cataractous changes in all 7000 ppm groups, with the severity ranging from minimal to marked: F_1 males - 4/30 treated vs 0/37 controls; F_1 females - 2/30 treated vs 0/40 controls; F_2 males - 11/33 treated vs 0/30 controls; F_2 females - 2/31 treated vs 0/32 controls. Minimal unilateral cataractous change was also observed in the 1500 ppm F_2 males (2/18 treated vs 0/30 controls).

Body weights were decreased in all 7000 ppm groups: F_1 males from PND 8 to weaning (↓6-24%); F_1 females from PND 15 to weaning (↓14-22%); F_2 males from PND 15 to weaning (↓9-20%); F_2 females from PND 22 to weaning (↓12-17%). Body weights were also decreased in 1500 ppm groups on PND 22 and 29 (↓6-14%). Body weight gains, as calculated by the reviewers, were decreased in both generations in the 7000 ppm groups (↓16-24%).

Plasma tyrosine levels were elevated in a dose related manner in all F_1 treatment groups (exposed in utero). A statistical assessment of these data was not presented by the study investigators. However, increases ranged from 269-586% of controls in males and from 159-542% of controls in females. In addition, plasma tyrosine levels were even more dramatically elevated in F_2 pups. At the 10 ppm level, male tyrosine levels were 794% of controls and females levels were 665% of controls. At the 7000 ppm level, male tyrosine levels were 5009% of controls and females were 3945% of control levels.

The LOAEL for offspring toxicity for males and females is 10 ppm (LDT) (equivalent to 2.1/2.4 mg/kg/day [M/F]) based on tyrosinemia at all dose levels in F_{2a} pups (exposed in utero). In addition, ocular discharge was observed at all dose levels in F_1 pups (and in nearly all F_2 pup dose groups) with a 0 incidence in controls and cataractous changes were observed histologically at the high dose level. An offspring toxicity NOAEL was not observed.

Even though a NOAEL for effects observed in offspring was not determined, this reproductive study in the mouse is determined to be **acceptable/guideline (§83-4(b), reproduction)** and satisfies the requirements for a multigenerational reproductive toxicity study in mice as per the Hazard Identification Assessment Review Committee (March 13, 2001).

4.5 Chronic Toxicity

Adequacy of data base for chronic toxicity: The data base for chronic toxicity is considered complete and no additional studies are required at this time. In the rodent studies, the males appeared to be more sensitive than the females and the male rats were more sensitive than the male mice. In mice dosed for up to one year, reduced body weight gain and food utilization were observed in the males at the highest dose tested (1000 mg/kg). Similar effects were observed in males dosed for up to 80 weeks (mouse oncogenicity study [MRID 44505028] presented in section 870.4200b). There were no treatment-related effects noted in the female mice in either study. The eye, liver, and kidney were the primary target organs of mesotrione in the rat. Corneal opacity, corneal vascularization, and/or keratitis, increased liver weights and hepatocyte fat vacuolation and increased kidney weights were observed at the LOAEL (approximately 0.48 mg/kg) in the males in the rat chronic study (2-year rat study [MRIDs 44505035 and 44505036] presented in section 870.4300). The eye lesions and signs of liver toxicity were observed in the females at >LOAEL. Additionally biologically significant reduced body weight gains were observed in both sexes at >LOAEL. Corneal and lenticular opacities were observed in the dog chronic study but only at the high-dose (600 mg/kg). At the LOAEL (10 mg/kg), tyrosinemia was observed in both sexes and an increased incidence of erythrophagocytosis in the mesenteric lymph nodes was observed in the females.

870.4100a (870.4300) Chronic Toxicity – Rodent

In this chronic oral toxicity study (MRID 44505026), mesotrione (ZA 1296; Batch No. P17; 96.8% a.i.) was administered continuously in the diet to 60 C57BL/10J$_f$CD-1 mice/sex/dose at nominal dose levels of 10, 50, 350, or 7000 ppm (equivalent to 1.5, 7.8, 56.2, or 1114.0 mg/kg/day in males and 2.1, 10.3, 72.4, and 1494.5 mg/kg/day in females) for 3, 6, or 12 months. Two control groups of 60 C57BL/10J$_f$CD-1 mice/sex (120/sex) were fed untreated diet for 3, 6, or 12 months. There were 20 treated mice/sex/group per interval.

Mortality, clinical observations, body weights, food consumption, ophthalmoscopic observations, hematological parameters, clinical chemistry parameters, urinalysis parameters,

organ weights, and gross and microscopic pathological findings were unaffected by the test substance.

Body weight gain in the 7000-ppm males was reduced overall by 17%. Reductions in body weight gain appeared to occur throughout the study. In conjunction with a 23% decrease in food utilization during the first 4 weeks of the study, an overall decrease in food utilization of 9% from weeks 1-13, and no decrease in food consumption throughout the study, the evidence suggests that there was a toxicologically significant effect in the 7000-ppm males.

Under the conditions of this study, there was no evidence of carcinogenic potential.

The LOAEL for this study is 7000 ppm (equivalent to 1114.0 mg/kg/day in males and 1494.5 mg/kg/day in females) based on decreases in body weight gain and food utilization in males. The NOAEL is 350 ppm (equivalent to 56.2 mg/kg/day in males and 72.4 mg/kg/day in females).

The submitted study is classified as **acceptable/guideline [§83-1(b)]** and satisfies the guideline requirements for a chronic oral toxicity study in rodents.

870.4100b Chronic Toxicity - Dog

In this chronic oral toxicity study (MRID 44505027), mesotrione (ZA1296; 97.6% a.i., Lot/batch # P22) was administered via gelatin capsule for 1 year to 4 Beagle dogs/sex/dose at concentrations of 0, 10, 100, or 600 mg/kg/day. Body weights, body weight gain, food consumption, hematology, clinical chemistry and urinalysis parameters, and organ weights were not adversely affected by the test substance.

At 10, 100, and 600 mg/kg/day, plasma tyrosine was dose-dependently increased ($p \leq 0.01$) relative to controls (males-↑168, 708, and 915%, respectively; females-↑151, 1213, and 1470%, respectively). During the clinical observations, cloudy eyes were observed in a single high-dose male (1/4). At the ophthalmological examination, lenticular opacities were noted in one high-dose male and one high-dose female. On gross pathology, one high-dose male had corneal and lenticular opacities with ocular discharge with a corresponding keratitis seen on microscopic evaluation. One high-dose female also had a corneal opacity with corneal erosion seen microscopically. A different high-dose male had red spots in the eye corresponding to peri-orbital hemorrhage. The corneal and lenticular effects are considered treatment related. No ocular effects were noted at the low and mid-doses. Interdigital cysts were observed in the mid- (males-2/4, females-1/4) and high- (males-2/4, females-3/4) dose animals and in the low-dose males (1/4) and control females (1/4). There also appeared to be an increase in dry skin sores in treated animals, especially males. These effects corresponded with an increase in staining of the hair, thorax, limbs, and paws, and green-colored urine as the dose increased. The skin effects were most likely due to the irritant properties of the phenolic acid metabolites in the urine that contacted the skin and may not be toxicologically relevant. Dermatitis/folliculitis was noted in the histopathology report in areas where dry sores or interdigital cysts occurred. Other areas that

were grossly normal showed no abnormalities. Minimal to moderate erythrophagocytosis in the mesenteric lymph nodes was observed during the histopathological examination in the 10 (3/4). 100 (2/4), and 600 (2/4) mg/kg/day females and in the 100 and 600 mg/kg/day males (1/4 and 2/4, respectively). This effect was considered treatment related.

At 600 mg/kg/day, one female was humanely killed during week 47 due to adverse clinical signs including convulsions, hypothermia, a slow, weak pulse, and rapid weight loss. Pathological examination of this animal indicated generalized lymphocytolysis. This death was considered treatment related.

The LOAEL for this study is 10 mg/kg/day based on evidence of tyrosinemia in both sexes and increased incidence of erythrophagocytosis in the mesenteric lymph nodes of females.

No NOAEL was determined for this study.

The submitted study is classified as **acceptable/guideline (§83-1b)** and satisfies the requirements for a chronic oral toxicity study in dogs.

4.6 Carcinogenicity

Adequacy of data base for Carcinogenicity: The data base for carcinogenicity is considered complete and no additional studies are required at this time. There is no evidence of carcinogenic potential in either the rat or mouse.

870.4200a Carcinogenicity Study - Rat

The 2-year feeding study in rats (MRIDs 44505035 and 44505036) presented in section 870.4300 satisfies the data requirements for 870.4200a.

870.4200b Carcinogenicity (feeding) - Mouse

In a mouse oncogenicity study (MRID 44505028), mesotrione (96.8% a.i., Lot/Batch # P17) was administered in the diet to C57BL/10J$_f$CD-1 Alpk mice (55/sex/group) for up to 80 weeks at 0, 10, 350, or 7000 ppm (equivalent to 0/0, 1.4/1.8, 49.7/63.5, and 897.7/1102.9 mg/kg/day [M/F], respectively). The high-dose animals received 3500 ppm of mesotrione for the first 7 weeks of the study and then received 7000 ppm for the remainder of the dosing interval.

The doses were selected on the basis of a 90-day feeding study in mouse carried out in the performing laboratory; no further information was provided. In a subchronic oral toxicity study (MRID 44505022) reviewed with the current submission, mesotrione (96.8% a.i) was administered for 13 weeks to 20 mice/sex/dose at dietary concentrations of 10, 50, 350, or 7000 ppm. All the parameters were unaffected. The NOAEL was 7000 ppm and no LOAEL was observed.

Mortality, clinical signs, food consumption, hematology, organ weights, and macroscopic and histopathological findings for both sexes at all doses were unaffected by treatment with mesotrione. In the 7000 ppm females and in both sexes of the 10 and 350 ppm dose groups, body weights, body weight gains, and food efficiency were also unaffected.

In the 7000 ppm males, slight, but consistent mean body weight reductions (↓2-9%; $p<0.05$ or 0.01) were observed during weeks 13 to 81. Overall (weeks 1 to 81) body weight gain (calculated by the reviewers) was reduced by approximately 20% compared to controls. Mean food consumption was consistently increased (↑4-10%; $p<0.01$) compared to controls during the first 12 weeks of the study and generally similar to controls thereafter, but food efficiency was reduced ($p<0.05$ or 0.01) during weeks 1-4 (↓12%), 9-12 (↓40%), and overall (weeks 1-12: ↓16%).

The LOAEL is 7000 ppm (equivalent to 897.7/1102.9 mg/kg/day M/F) based on minimal but, consistently reduced body weights, and reduced body weight gains and food efficiency in males. The NOAEL is 350 ppm (equivalent to 49.7/63.5 mg/kg/day M/F).

In this study no treatment-related neoplastic changes were observed.

The submitted study is classified as **acceptable/guideline (§83-2b)** and does satisfy the guideline requirements for a carcinogenicity study in mice.

870.4300 Chronic/Oncogenicity - Rat

In a combined chronic/oncogenicity study (MRID 44505035, 44505036), mesotrione (96.8% a.i.) was administered via the diet to 64 Alpk:APfSD rats/sex/group at 0, 7.5, 100, or 2500 ppm (equivalent to 0, 0.48, 6.48 or 159.89 mg/kg/day in males and 0, 0.57, 7.68, or 189.48 mg/kg/day in females) for up to 104 weeks. To assess ocular toxicity, an additional 20 rats/sex/dose were dosed at 1 or 2.5 ppm (equivalent to 0.06 and 0.16 mg/kg/day in males and 0.08 and 0.19 in females). Twelve main study rats/sex/dose were terminated after 52 weeks.

No treatment-related adverse effects were observed on mortality, food consumption, food efficiency, or hematology, clinical chemistry, and urinalysis parameters for either sex at any treatment level. All male groups were terminated when survival dropped to approximately 25% during weeks 92/93 and 97/98. Chronic progressive glomerulonephropathy was the major contributory factor involved in the intercurrent deaths. There was no evidence of a trend in male or female Kaplan-Meier survival rates. Female groups were terminated as scheduled in week 104.

The major target organ was the eye. In the 7.5-, 100-, and 2500-ppm males and in the 100- and 2500-ppm females ocular lesions consisting of cloudy eyes, corneal lesions consisting of opacity/hazy opacity/vascularization/ghost vascularization, and/or corneal keratitis were

observed. No treatment-related ocular lesions were observed in the 1- and 2.5-ppm animals or in females dosed at 7.5 ppm.

Reductions ($p<0.05$ or 0.01) in mean body weight were observed throughout the study in the 7.5-ppm males (↓2-13%) and the 100- and 2500-ppm males and females (↓1-17%). Body weight reductions ($p<0.05$ or 0.01) were also observed in the 2.5- and 1-ppm male satellite groups. Body weight decreases of over 10% in the 2.5- and 7.5-ppm males were noted only from week 75 to termination after body weights had stabilized in all groups. Since there were random fluctuations of the weights among all groups during this time period, body weight changes at 7.5 ppm and below were not considered biologically relevant. Decreases in body weight gain from weeks 1 to 47 were only biologically significant in males at 100 and 2500 ppm and, in females, at 2500 ppm. There was no dose relationship with the decrease in body weight gain in either sex among all treatment groups from week 47 to termination.

Increases ($p<0.01$) in absolute kidney weights in the 7.5-ppm and the 2500-ppm males (↑12-15%) and adjusted (for body weight) kidney weights in the 7.5-, 100-, and 2500-ppm males (↑13-22%) were observed at the interim sacrifice (week 53). These differences in kidney weights were not observed at the terminal sacrifice.

Increases ($p<0.05$ or 0.01) in absolute and adjusted liver weights in the 7.5-ppm males (↑18 and 17%, respectively), adjusted liver weights in the 100- and 2500-ppm males (↑15-18%), and adjusted liver weights in the 100- and 2500-ppm females (↑11-14%) were observed at the interim sacrifice. At the terminal sacrifice, increases ($p<0.05$ or 0.01) in absolute and adjusted liver weights in the 100- and 2500-ppm males (↑7-20%) were observed. During gross examination of the 7.5-, 100-, and 2500-ppm dose groups, pale liver was observed in the males (25-27/treated group vs. 9 controls) and females (3-10/treated group vs. 2 controls). During histopathological examination, minimal to marked hepatocyte fat vacuolation was observed in the 7.5-, 100-, and 2500-ppm males (36-39/treated group vs. 17 controls) and the 100- and 2500-ppm females (14-16/treated group vs. 8 controls).

Decreased absolute adrenal weights in the 100- and 2500-ppm males (↓21 and 22%, respectively) and decreased adjusted adrenal weights in the 7.5-, 100-, and 2500-ppm males (↓19, 25, and 29%, respectively) showed a slight dose-dependent trend, but no histopathological abnormalities were observed.

The administration of mesotrione to rats up to 2500 ppm (159.9 mg/kg/day for males, 189.5 mg/kg/day for females) in the diet did not result in an overall treatment-related increase in incidence of tumor formation.

Under the conditions of this study, dosing is considered adequate to assess the carcinogenic potential of mesotrione based on the ocular and hepatic lesions and increased liver and kidney weights noted at 7.5 ppm and above and body weight effects at 100 ppm and above in males and increased liver weights and ocular and hepatic lesions at 100 ppm and above in females.

The LOAEL for this combined chronic toxicity/ carcinogenicity rat feeding study is 7.5 ppm (0.48 mg/kg/day for males, 0.57 for females) based on ocular lesions, increases in kidney and liver weights, and hepatocyte fat vacuolation in males. No NOAEL was determined for kidney and liver weights or hepatocyte fat vacuolation in males. The NOAEL for ocular lesions in the special study is 2.5 ppm (0.16 mg/kg/day for males, 0.19 mg/kg/day for females).

The submitted study is classified as **acceptable/guideline (§83-5)** and does satisfy the requirements for a chronic toxicity study (§83-1) and a carcinogenicity study (§83-2) in rats.

4.7 Mutagenicity

Adequacy of data base for Mutagenicity: The data base for mutagenicity is considered adequate based on 1991 mutagenicity guidelines. The genetic toxicology studies indicate that mesotrione was not mutagenic in *Salmonella typhimurium*, *Escherichia coli* or in cultured mouse lymphoma cells. There was also no evidence of clastogenicity *in vitro*, and mesotrione gave a negative response for the induction of micronucleated polychromatic erythrocytes in bone marrow. Reverse gene mutation assays also indicated that the mesotrione metabolites MNBA and AMBA are not mutagenic. Based on the available mutagenicity studies, there is no concern for mutagenic potential.

Gene Mutation-Mesotrione (ZA 1296)

870.5100- reverse gene mutation assay in bacteria MRID 44373526 Acceptable	*S. typhimurium* and *E. coli* were exposed to mesotrione (ZA 1296) at concentrations of 100 to 5000 μg/plate ± S9. There was *no evidence* of induced mutant colonies over background.
870.5300-gene mutation assay in mouse lymphoma cells MRID 44373525 Acceptable	L5178Y TK +/- mouse lymphoma cells cultured *in vitro* were exposed to ZA 1296 at concentrations ranging from 125 to 1000 μg/mL ± S9. ZA 1296 was negative for inducing forward mutations at the TK locus.

Cytogenetics-Mesotrione (ZA 1296)

870.5375, *in vitro* mammalian cytogenetics assay MRID 44373524 Acceptable	Human lymphocytes were exposed to ZA 1296 at 0, 250, 1000, 1500, or 2000 μg/mL (-S9) and at 0, 250, 1000, and 2000 μg/mL (+S9). ZA 1296 was not clastogenic with S9 activation and was equivocal for clastogenic activity without S9 activation.
870.5395, bone marrow micronucleus assay MRID 44373527 Acceptable	CD-1 mice were treated by gavage with ZA 1296 at single dose of 500 mg/kg. ZA 1296 gave a negative response for the induction of micronucleated polychromatic erythrocytes in bone marrow at both sampling times.

Gene Mutation-Mesotrione Metabolites (MNBA and AMBA)

870.5100- reverse gene mutation assay in bacteria MRID 44505037 Unacceptable/not upgradable	*S. typhimurium* and *E. coli* were exposed to MNBA at concentrations of 100 to 5000 μg/plate ± S9. There was *no evidence* of induced mutant colonies over background except for an equivocal response with tester strain TA100 at 5000 μg/plate (+S9).
870.5100- reverse gene mutation assay in bacteria MRID 44505038 Acceptable	*S. typhimurium* and *E. coli* were exposed to AMBA at concentrations of 100 to 5000 μg/plate ± S9. There was *no evidence* of induced mutant colonies over background.

4.8 Neurotoxicity

Adequacy of data base for Neurotoxicity: The data base for neurotoxicity is not complete. The HIARC recommended that a developmental neurotoxicity study be required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects. There is some concern about the effects of elevated plasma tyrosine levels on the developing nervous system in children due to a report by Ruetschi *et al* (2000) that some patients with tyrosinemia III (an autosomal recessive disorder in which 4-hydroxyphenylpyruvate dioxygenase is deficient) were presented with mental retardation or neurological symptoms; no correlation of the severity of the mutation and enzyme deficiency and mental function has been found. Also, tyrosine levels did not correlate with the clinical phenotype. There was no evidence of neurotoxicity or neuropathology observed in the acute and subchronic neurotoxicity studies. In the multi-generation mouse reproduction study, one F_1 male and one F_1 female had retinal detachment with marked cataractous changes at the highest dose tested (>1000 mg/kg). In the subchronic toxicity dog study, the high-dose females had decreased absolute and relative brain weights; however, no microscopic abnormalities were noted in any brain tissue from the high-dose group and the effect was not observed in the chronic toxicity dog study.

870.6200 Acute Neurotoxicity Screening Battery

In this acute oral neurotoxicity study (MRID 44505017 and 44505018), mesotrione (ZA1296; 97.6% a.i., Lot/batch # P22) was administered in a single dose by gavage to 10 Alpk:AP$_f$SD rats/sex/dose at doses of 0, 20, 200, or 2000 mg/kg. After two weeks, five animals/sex/group were perfused for neurohistological examination and animals from the control and high-dose groups were examined microscopically. Functional observational battery (FOB) and motor activity were evaluated during week -1 and on days 1 (approximately 2 hours post-dosing), 8, and 15. No treatment-related deaths occurred. Clinical signs, FOB parameters, motor activity, body weights, body weight gains, food consumption, gross pathology, histopathology, and brain weights and dimensions were unaffected by the test substance.

The LOAEL is >2000 mg/kg, based upon lack of any systemic effects. The NOAEL for this study is 2000 mg/kg.

Although no effects were noted at the high dose, the substance was tested at the limit dose, so the high dose is acceptable. This study is classified as **acceptable/guideline** and satisfies the requirements for an acute neurotoxicity screening battery in rats (§81-8a; 870.6200).

870.6200 Subchronic Neurotoxicity Screening Battery

In this subchronic neurotoxicity screening battery, mesotrione (ZA1296, 97.6% a.i., Lot/Batch P22) (MRID 44505025) was administered continuously in the diet for 90 days to 12 Alpk:AP$_f$SD rats/sex/group at doses of 0, 2.5, 100, or 5000 ppm (equivalent to [M/F] 0/0, 0.20/0.23, 8.25/9.29, or 402.8/466.6 mg/kg/day). Five animals/sex/group were perfused for neurohistological examination and animals from the control and high-dose groups were examined microscopically. The functional observational battery (FOB) and motor activity were evaluated during weeks -1, 5, 9, and 14.

No treatment-related deaths occurred. Food consumption and utilization, FOB parameters, motor activity, brain dimensions, and neuropathology were unaffected by the test substance.

Corneal opacities and/or vascularization of the cornea were seen in 3/11 mid-dose males and 1/12 mid-dose females. Corneal opacities and/or vascularization of the cornea were seen in 10/12 high dose males, and 7/12 high dose females. Overall (weeks 1-14) body weight gains were decreased in the high-dose females (↓18%). High-dose females displayed decreased body weights (adjusted for initial body weight) from week 2 until study termination (↓5-9%).in the high-dose females at week 14 (↑37%). No treatment-related findings were observed in the 2.5 ppm group.

The LOAEL for this study is 100 ppm (equivalent to 8.25 mg/kg/day in males and 9.29 mg/kg/day in females) based on corneal opacities and/or vascularization of the cornea of the eye. The NOAEL for this study is 2.5 ppm (equivalent to 0.20 mg/kg/day in males and 0.23 mg/kg/day in females).

The submitted study is classified as **acceptable/guideline (§82-7[a])** and satisfies the guideline requirements for a subchronic neurotoxicity screening battery in rats.

870.6300 Developmental Neurotoxicity Study

The HIARC recommended that a developmental neurotoxicity study be required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects.

4.9 Metabolism

Adequacy of data base for metabolism: The data base for metabolism is considered complete. A series of rat metabolism studies with [^{14}C-aromatic]mesotrione indicated that mesotrione was readily absorbed and distributed in the body. Tissue distribution was about the same in both sexes, although one study showed higher residues in the kidneys in females, with the highest residues of the test compound in the liver and kidney. Higher doses resulted in higher residues in the liver and kidney, while repeated doses resulted in reduced accumulation of residues in all tissues. Levels of radioactivity in tissues of iv-dosed animals were essentially the same as in orally-dosed animals. Over 50% of the administered dose was excreted in the urine in both sexes and around 25% was excreted in the feces within 72 hours. Females exhibited slightly higher total urinary excretion than males, but total fecal excretion was about the same in both sexes. Increasing the dose or repeated doses had little effect on the pattern of excretion in both sexes. The overall pattern of excretion was similar between orally-dosed and iv-dosed rats. The metabolite profile was similar between the sexes in each group and between the single-dosed and repeated-dosed animals. The parent compound, mesotrione, was the major component identified in the urine accounting for 47-64% of the dose. In addition, the following minor metabolites were identified: MNBA (1-4% of the dose), AMBA (3-12%), 5-hydroxymesotrione ($\leq$ 2%), and 4-hydroxymesotrione (3-6%). In bile cannulated rats administered [^{14}C-aromatic]mesotrione or [^{14}C-dione]mesotrione, the major component in fecal excreta and bile was the parent compound. Analysis of the bile identified mesotrione and 4-hydroxymesotrione as two minor components. Another minor component in the feces was 5-hydroxymesotrione. Metabolism in the mouse was very similar to the rat except that males had slightly increased total fecal excretion when compared to females and, females in the low-dose group excreted higher (1.5x) levels of parent compound in the urine than males. Free mesotrione was the major component in the urine and feces ($\geq$ 50% of the dose). Minor components in the fecal extracts included AMBA (1-4%) and MNBA ($\leq$ 2%).

870.7485 Metabolism - Rodent

1) In a series of rat metabolism studies (MRIDs 44505101 through 44505106), [^{14}C-aromatic]mesotrione ($\geq$98.1% radiochemical purity) was administered to Alpk:AP$_f$SD rats (5/sex/dose) as either a single oral (gavage) dose at 1.00 or 100 mg/kg, a single intravenous (iv) dose at 1.00 mg/kg, or a single oral dose at 1.00 mg/kg following a 14-day pretreatment with mesotrione at 1.00 mg/kg/day. In addition, 2 bile-duct cannulated rats/sex were administered a single oral dose of [^{14}C-aromatic]mesotrione at 50.0 mg/kg or a single oral dose of [^{14}C-dione]mesotrione ($\geq$99% radiochemical purity) at 50.0 mg/kg.

The overall recovery of dosed radioactivity in excreta, bile, tissues, cage washes was 92.0-97.1% from rats in the mass balance studies and 62.5-92.9% from rats in the biliary excretion study. Within 72 hours of receiving a single oral dose of [^{14}C-aromatic]mesotrione at 1.00 mg/kg, both sexes excreted 54.2-55.9% of the dose in the urine and 23.8-24.5% of the dose in the feces. Radioactivity remaining in the carcass/tissues of both sexes accounted for 11.2-12.5% of the dose. Increasing the dose to 100 mg/kg had little effect on the pattern of excretion with both

sexes. Males and females still excreted the majority of the dose in the urine (61.5-63.0% dose), with fecal excretion accounting for 28.8-30.5% dose, although the recovery of radioactivity in the tissues/carcass was lower (0.71-1.1% dose). Repeated dosing at 1.00 mg/kg/day also had little effect on the pattern of excretion; although the levels of urinary (60.8-67.0% dose) and fecal (23.1-30.3% dose) excretion were slightly increased, and recovery of radioactivity in the tissues/carcass was lower (5.1-5.3% dose). The overall pattern of excretion was also generally similar between rats dosed orally or intravenously at 1.00 mg/kg. Radioactivity in urine of the iv-dosed males (79.4% dose) and females (84.1% dose) was somewhat higher (1.5x) than in urine of orally dosed rats, and iv-dosed rats had lower (0.10-0.28x) levels of radioactivity in feces (2.4-6.8% dose). However, intravenous dosing resulted in similar levels of radioactivity being retained in the tissues and carcass (10.0-10.4% dose) after 72 hours.

In the bile-duct cannulated rats administered [^{14}C-aromatic]mesotrione, a similar pattern of elimination was noted between the sexes with the majority of the administered dose recovered in the urine; further, urinary excretion was slightly higher (1.2x) in females (64.1% dose) than in males (55.2% dose). Radioactivity in the feces accounted for 25.3 and 26.8% of the dose in males and females, respectively, while biliary excretion was a minor route of excretion for males (10.4% dose), but even lower (0.19x) for females (2.0% dose). Altering the position of the ^{14}C-label within the parent molecule from the aromatic-ring to the dione-ring had only a minor impact on the pattern of excretion in rats. Approximately 46% of the dose was excreted in the urine and approximately 13% of the dose excreted in the feces in both males and females. The recovery of radioactivity decreased (0.4-0.8x) slightly in the urine (44.1-47.5% of dose) and feces (11.2-16.2% dose) of males and females. Biliary excretion of bile-duct cannulated rats was a minor route of excretion for males (14.2% dose), and was even lower (0.27x) for females (3.8% dose).

Concentrations of ^{14}C-residues in tissues were similar between the sexes within each dose group. Although actual ^{14}C-residue concentrations in tissues differed between dose groups, the relative distribution of radioactivity between tissues was the same within each dose group, with ^{14}C-residues being highest in liver and kidney. In each low-dose group, radioactivity in the female kidneys was 3.3-4.4x higher than males; in the high-dose group, the difference was 1.8x. Increasing the dose level from 1.00 to 100 mg/kg increased the concentration of radioactivity in liver (1.9-2.1x) and kidneys (1.5-3.0x) in both sexes. Repeated dosing resulted in reduced accumulation of ^{14}C-residues in tissues, with ^{14}C-residues being 0.44x lower on average in tissues of repeated dose animals. Levels of radioactivity in tissues of iv dosed animals were essentially the same as in orally dosed animals.

With the exception of the bile-duct cannulated group of rats, 62-78% of the dose was identified in urine and fecal extracts of orally dosed rats. Although there were minor differences in levels of metabolites between males and females and between dose groups, the metabolite profile was similar between the sexes in each group and between the single low-dose, single high-dose, and repeated low-dose groups. In rats from each of these groups, free mesotrione was the major metabolite identified in urine, accounting for 47-64% of the dose. In addition, the following minor metabolites were identified: 2-nitro-4-(methylsulphonyl)-benzoic acid (MNBA, 1-4%

dose), 4-(methylsulphonyl)-2-aminobenzoic acid (AMBA, 3-12% dose), 5-hydroxy-mesotrione (≤2% dose), and 4-hydroxy-mesotrione (3-6% dose).

In bile cannulated rats administered a 50 mg/kg dose of [^{14}C-aromatic]mesotrione, a total of 74-77% of the dose was identified in excreta and bile, with the major component being parent compound. Analysis of bile identified only two minor components, 4-hydroxy mesotrione (2% dose, males only) and mesotrione (2-8% dose). Although biliary excretion was a minor route of elimination for both sexes, it was more prominent in males (10% dose) than females (2% dose). For both sexes, the majority of the radioactivity was excreted in the urine and was identified as parent (males, 48% dose; females, 60% dose). AMBA was the major component in feces and accounted for 8-10% of the dose.

In bile cannulated rats administered a 50 mg/kg dose of [^{14}C-dione]mesotrione, a total of 55-59% of the dose was identified in excreta and bile. Analysis of bile identified only two components, 4-hydroxy mesotrione (1% dose, males only) and parent (3-11% dose). Biliary excretion was again a more prominent route of excretion for males (12% dose) than females (3% dose). For both sexes, the most prominent route of excretion was in the urine and the major component identified in urine was parent, totaling 43 and 51% of the dose in males and females, respectively. Two minor components were identified in the fecal extracts, 5-hydroxy mesotrione (1% dose, females only) and parent (1% dose, males only).

This is study is classified **acceptable (§85-1a)** and does satisfy the guideline requirements for a metabolism study in rats.

2) In this mouse metabolism study (MRID 44537101), [^{14}C-aromatic]mesotrione (97% radiochemical purity) was administered to CD-1:Crl(ICR)BR mice (4/sex/dose) as a single oral (gavage) dose at 1.00 or 100 mg/kg.

Absorption of [^{14}C-aromatic]mesotrione from the G.I. tract of mice was evident in low- and high-dose animals based upon the high level of urinary excretion. In both dose groups, overall renal and fecal excretion accounted for 67.27-90.94% of the dose within 24 hours of dosing, equivalent to 86-96% of the total excretion. The pattern of excretion was different for the low-dose sexes, but was similar between the high-dose sexes.

At the low-dose level, females exhibited higher total urinary excretion (1.4x) when compared to the males and males had increased total fecal excretion (1.8x) compared to the females. No further differences were noted in the low-dose sexes in levels of radioactivity recovered in the terminal cage wash, GI contents, or tissues/carcass. At the high-dose, male and female mice excreted 62.90-69.82% of the dose in the urine and 24.46-27.27% of the dose in the feces within 72 hours of administration. Radioactivity remaining in the carcass/tissues of both sexes from the high-dose group accounted for 0.28-0.41% of the dose. When compared to the low-dose mice, greater elimination occurred in the urine of the high-dose animals (1.2-1.5x). Additionally, radioactivity recovered in the tissues/carcass of high-dose animals was much lower (0.019-

0.029x) in comparison to the low-dose animals and may indicate that bioaccumulation in the tissues may have been saturated at the high-dose level.

At 72 hours post-dose, ^{14}C-residues were highest in liver and kidney and lowest in heart, lung, muscle, bone, brain, and plasma. In low- and high-dose females, the higher concentration of radioactivity in the kidneys corresponded to the higher level of urinary excretion.

Except for the kidneys, levels of radioactivity in tissues and organs were similar between sexes 72 hours following a low-dose at 1.00 mg/kg. ^{14}C-residues levels in the kidneys were 4.3x higher in the females when compared to the males; this difference was the single sex-related variation. In the high-dose group, females had higher levels of radioactivity in the liver (1.7x), kidneys (5.7x), and fat (5.1x) when compared to males. Increasing the dose level from 1.00 to 100 mg/kg increased the concentration of radioactivity in tissues by 25.8x on average for both sexes, with the greatest increases occurring in fat (5.7-65x), carcass (11-46x), and whole blood (22-104x).

For metabolite characterization, 51-81% of the dose was identified in urine and fecal extracts. In mice from both the low- and high-dose group, free mesotrione was the major component identified in urine and feces, accounting for 49-65% of the dose in the low-dose group and 70-78% of the dose in the high-dose group. In low- and high-dose mice, minor components detected in fecal extracts included 4-(methylsulphonyl)-2-aminobenzoic acid (AMBA, 1-4% dose) and 2-nitro-4-(methylsulphonyl)-benzoic acid (MNBA, ≤2% dose). No urinary or fecal unknowns were detected which accounted for >5% of the administered dose.

When the high-dose mice were compared to the low-dose group, the metabolite profile was qualitatively similar. The only difference observed was in the low-dose group, in which the females excreted higher (1.5x) levels of parent compound in the urine than males.

In low-dose female mice, increased renal excretion of parent compound suggested a more efficient excretion route for the females. No apparent sex-related differences were noted in the rat metabolism studies submitted with the current study (MRIDs 44505101 through 44505106) regarding patterns of elimination or the metabolic profile.

This study is classified **acceptable (§85-1)** and does satisfy the requirement for a metabolism study.

870.7600 Dermal Absorption - Rat

There are no dermal absorption studies available.

Special Studies for Ocular Effects - Subchronic Oral - Rat

A summary of the conclusions from the special studies for ocular effects is presented at the end of this section as part of the executive summary of a study in which 50 triketone compounds

were evaluated to assess the relationship between plasma and ocular tyrosine levels and between plasma tyrosine and the incidence of corneal lesions in the rat.

1) In a non-guideline study (MRID 44537104), mesotrione (96.8% a.i.; lot # P17) was administered in the diet at 2500 ppm (equivalent to 272 mg/kg/day) to Alpk:AP$_f$SD male rats (40 rats treated and 16 control rats) for 5 weeks. Any ocular changes in the treated animals were graded. At the end of the treatment period and, all animals were allocated (based on the severity of any ocular lesions) to receive a gross postmortem examination or to undergo an 8 week recovery period.

No mortalities occurred during the study. When compared to concurrent controls, no treatment-related changes were observed in body weights, body weight gains (calculated by reviewers), food consumption, food utilization, or gross pathology.

During clinical observation, cloudy eye(s) were observed in 22/40 treated males; this finding was not observed after week 11. In the treated animals, the ophthalmoscopic examination at week 1 revealed slight corneal opacity (2/80 examined eyes); by week 2, the incidence of this finding had increased (14/80 eyes). During week 3, slight to moderate corneal opacities (24/80 eyes) were observed. During weeks 4 and 5, slight to marked corneal opacities (34 and 39/80 eyes, respectively) and vascularization (3 and 7/80 eyes, respectively) were observed. At week 6 (week 1 of recovery), slight to moderate corneal opacities (11/30 eyes examined), slight to marked hazy opacities (7/30 eyes), and vascularization (9/30 eyes) were noted. During week 7, slight corneal opacity (1/30 eyes), marked hazy opacity (1/30 eyes), vascularization (6/30 eyes), and ghost vascularization (9/30 eyes) were observed. At week 10, 1/30 eyes showed vascularization, while 11/30 eyes displayed ghost vascularization; during week 12, ghost vascularization was noted in 11/30 eyes. Histological abnormalities of the cornea were observed in treated males. No abnormalities were noted in the control group. At the end of treatment, the following observations were noted in the eyes of the treated males: (i) minimal (1/25 animals examined), slight (6/25), or moderate (4/25) keratitis; and (ii) minimal (5/25), slight (4/25), or moderate (1/25) polymorphonuclear leukocytic infiltration at filtration angle. At the end of the recovery, the following observations were noted in the eyes of the treated males: (i) minimal (6/15 animals examined) or slight (2/15) corneal vessels; (ii) minimal (6/15) or slight (2/15) corneal stromal fibroblasts; and (iii) minimal (2/15) or slight (1/15) corneal epithelial disruption.

When compared to concurrent controls, plasma tyrosine levels were decreased ($p \leq 0.01$) during week 1 (↓47%) and increased ($p \leq 0.01$) during weeks 2 (↑1967%), 6 (↑1419%), 7 (↑1254%), and 14 (↑16%).

In conclusion, corneal lesions associated with dietary administration of 2500 ppm mesotrione for 5 weeks were resolved ophthalmoscopically and histologically following an 8 week recovery period.

This special study is classified as **acceptable/non-guideline** and satisfies the purpose for which is was intended.

2) The objective of this study (MRID 44537105) was to investigate ocular lesions in rats fed tyrosine in the diet. Alpk:AP$_f$SD weanling rats (21-23 days old; 8/group) were fed a low protein diet (PCD low protein diet) supplemented with 0, 0.5, 1.0, 2.5, or 5% (w/w) L-tyrosine (100% purity assumed) for 21 days. Ophthalmoscopic examinations were performed during acclimatization, daily on days 2 through 8, and on days 11, 12, 14, 18, and 21. Upon detection of corneal lesions in the majority of the rats in a group, the group was terminated. All remaining groups were terminated on day 21. All animals were necropsied and the eyes and Harderian glands examined by light microscopy.

Corneal lesions were first detected on day 3 in the 5.0% group (5/8 rats) and by day 4 (2/8 rats) in the 2.5% tyrosine group. By days 4 and 6, respectively, 6/8 rats in each group were affected. The lesions started as single or multiple small foci of opacity, which with time coalesced to produce larger areas of opacity. Fixation of the iris, indicating iritis, was observed in both the 2.5 and 5.0% groups (1/8 rats, each). Histopathological examination detected minimal/slight keratitis in both effected groups (4/8 rats, each); the keratitis was characterized by polymorphonuclear leucocyte infiltration of the corneal outer stroma and focal corneal epithelium disorganization. Minimal epithelial disorganization without inflammation was observed in 2/8 rats fed 5% tyrosine. The results of this special study are presented as an attachment to this overview (study report Tables 2 and 3, pages 10 and 11).

In conclusion, tyrosine at levels of 2.5 and 5% in a low protein diet induces ocular toxicity in Alpk:AP$_f$SD weanling rats.

3) The objective of this study (MRID 44505110) was to compile the results of 15 studies in which over 50 triketone compounds were evaluated to assess the relationship between plasma and ocular tyrosine levels and between plasma tyrosine and the incidence of corneal lesions in the Alpk:Ap$_f$SD rat. Each study consisted of 1 negative control group (typically 8 males), one positive control group (typically 12 males treated with SC0735 or ICIA0051), and up to 6 groups (typically 16 males/group) treated with one of the triketone analogues. Exposure periods ranged from 4 to 13 weeks.

The studies compiled in this report were designed to evaluate the ocular toxicity of a group of structurally divergent triketones in the rat and to identify those structural features of a triketone which affect its ocular toxicity potential. The identities of the tested compounds and the details of the study designs are presented in an attachment (Study report Tables 1 and 2, pages 50-54). In the initial studies, rats were dosed orally by gavage with triketones (10 mg/kg, nominally) daily for up to 6 weeks. In the later studies, dietary dosing was conducted and test substances were administered at 1 to 80 ppm daily for 4-13 weeks. The NOAEL for ocular toxicity induced by ICIA0051, the positive control used for the dietary studies, has been defined in a 2 year rat bioassay as 1 ppm; therefore, comparison of the ocular toxicity of novel triketones with ICA0051 over a 6 week period was expected to give an indication of the relative ocular toxicity potential of these compounds in a 2 year bioassay. Ophthalmoscopic examinations were conducted before

and during the study, and at study termination. In addition, plasma and ocular tyrosine measurements were made during the study and/or at study termination.

The following conclusions were drawn from these gavage and dietary studies of triketones in the rat:

- Many triketones caused increased plasma and ocular tyrosine concentrations and induced ocular lesions.
- Structurally similar triketones can have markedly different potencies.
- The least potent test substances included enamine pro-herbicide derivatives of triketones.
- Plasma and ocular tyrosine concentrations are closely related; ocular tyrosine levels are generally twice as high as plasma tyrosine levels
- Higher plasma tyrosine concentrations increased the risk of corneal lesions
- Evidence suggests that there may be a tyrosine threshold (1000 nmol/mL in plasma) for ocular lesion development.
- The ocular lesion produced by triketones is due to prolonged tyrosinemia rather than a direct effect of the triketone on the cornea.

4.10 Special/Other Studies

Special/range-finding rodent, human and metabolite studies are summarized in the subchronic and prenatal developmental toxicity sections of this Chapter.

5.0 HAZARD ENDPOINT SELECTION

On March 13, 2001, the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) reviewed the recommendations of the toxicology reviewer for mesotrione with regard to the acute and chronic Reference Doses (RfDs) and the toxicological endpoint selection for use as appropriate in occupational/residential exposure risk assessments. The potential for increased susceptibility of infants and children from exposure to mesotrione was also evaluated as required by the Food Quality Protection Act (FQPA) of 1996. The Health Effects Division (HED) Mechanism of Toxicity Science Assessment Review Committee convened on January 31, 2001 to evaluate the mechanism of toxicity of mesotrione in rats and mice in regards to tyrosine-mediated effects and assess the relevance of the two animal models to human health. The Mechanism of Toxicity Committee determined that the petitioner has adequately demonstrated that for tyrosine-mediated toxicological effects, the mouse is a more appropriate model for assessing human risk than is the rat. This decision is based on comparative data on the activity of tyrosine aminotransferase (TAT) in the rat, mouse and human, and the similarities of the response to elevated plasma tyrosine levels in humans and the mouse (D272633; March 27, 2001).

5.1 See Section 9.2 for Endpoint Selection Table.

5.2 Dermal Absorption

Dermal Absorption Factor: No dermal absorption study was submitted. Using a ratio of the maternal LOAEL from the developmental rabbit study and the NOAEL from the rabbit dermal toxicity study, one can derive a dermal absorption factor of 25% as an upper-bound estimate.

$$\frac{\text{Dev. Rabbit LOAEL}}{\text{Dermal Tox. NOAEL}} = \frac{250 \text{ mg/kg/day}}{1000 \text{ mg/kg/day}} = 25\%$$

The dermal absorption factor is required for short-, intermediate- and long-term dermal risk assessment since oral doses were selected for these exposure periods.

5.3 Classification of Carcinogenic Potential

5.3.1 Conclusions

In accordance with the EPA Draft Guidelines for Carcinogen Risk Assessment (July, 1999), the HIARC classified mesotrione as "not likely to be carcinogenic to humans" by all routes of exposure based upon lack of evidence of carcinogenicity in rats and mice.

5.3.2 Classification of Carcinogenic Potential

Not likely to be carcinogenic to humans

5.3.2 Quantification of Carcinogenic Potential

Not applicable

6.0 FQPA CONSIDERATIONS

6.1 Special Sensitivity to Infants and Children

There is quantitative and qualitative evidence of increased susceptibility of rat, mouse and rabbit fetuses *in utero* in developmental studies.

There is quantitative and qualitative evidence of increased susceptibility of mouse offspring in the multi-generation reproduction study. The rat multi-generation reproduction study did not establish a NOAEL for parental or offspring systemic toxicity, but there was no qualitative evidence of increased susceptibility in the offspring; increased plasma tyrosine levels were seen in the parental animals and the F_2 pups.

6.2 Recommendation for a Developmental Neurotoxicity Study

The HIARC recommended that a developmental neurotoxicity study in the mouse be required in order to better characterize the effects of tyrosinemia on the developing nervous system and to correlate plasma tyrosine levels to neurotoxic effects.

7.0 OTHER ISSUES

The Mechanism of Toxicity Committee determined that the petitioner has adequately demonstrated that for tyrosine-mediated toxicological effects, the mouse is a more appropriate model for assessing human risk than is the rat. This decision is based on comparative data on the activity of TAT in the rat, mouse and human, and the similarities of the response to elevated plasma tyrosine levels in humans and the mouse.

8.0 REFERENCES

44373512 Robinson, P. (1994) ZA1296: acute oral toxicity to the rat. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4502, Study No. AR5800. November 4, 1994. Unpublished.

44373513 Lees, D., and H. Connolly (1997) ZA1296: acute oral toxicity to the rat of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4809, Study No. AR6121. August 8, 1997. Unpublished.

44373514 Robinson, P. (1994) ZA1296: acute dermal toxicity to the rat. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4503, Study No. CR3171. November 4, 1994. Unpublished.

44373515 Lees, D., and H. Connolly (1995) ZA1296: acute dermal toxicity to the rat of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4810, Study No. CR3278. November 29, 1995. Unpublished.

44373516 Rattray, N. (1995) ZA1296: 4-hour acute inhalation toxicity study in the rat. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4739, Study No. HR2273. August 8, 1995. Unpublished.

44373517 Parr-Dobrzanski, R. (1995) ZA1296: 4-hour acute inhalation toxicity study in the rat of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4822, Study No. HR2275. November 29, 1995. Unpublished.

44373518 Robinson, P. (1994) ZA1296: eye irritation to the rabbit. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4505, Study No. FB4997. November 3, 1994. Unpublished.

44373519 Lees, D., and H. Connolly (1995) ZA1296: eye irritation to the rabbit of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4804, Study No. FB5246. November 29, 1995. Unpublished.

44373520 Robinson, P. (1994) ZA1296: skin irritation to the rabbit. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4504, Study No. EB4314. November 4, 1994. Unpublished.

44373521 Lees, D., and H. Connolly (1995) ZA1296: skin irritation to the rabbit of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4803, Study No. EB4421. November 29, 1995. Unpublished.

44373522 Robinson, P. (1994) ZA1296: skin sensitization to the guinea pig. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4506, Study No. GG6226. November 2, 1994. Unpublished.

44373523 Lees, D., and H. Connolly (1995) ZA1296: skin sensitization to the guinea pig of a 480 g/L SC formulation. Zeneca Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4817, Study No. GG6556. November 29, 1995. Unpublished.

44373524 Griffiths, K., Mackay, J.M. (1994) ZA 1296: An Evaluation in the In Vitro Cytogenetic Assay in Human Lymphocytes. Zeneca Central Toxicology Laboratory (CTL), Alderley Park, Macclesfield, Cheshire, UK. Laboratory Project Report No. CTL/P/4187. Laboratory Project Study No. SV0700. February 21, 1994. Unpublished.

44373525 Clay, P. (1994). ZA 1296: Assessment of Mutagenic Potential Using L5178Y Mouse Lymphoma Cells. Zeneca Central Toxicology Laboratory (CTL), Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4183. Laboratory Study No. VV0098. February 28, 1994. Unpublished.

44373526 Callander, R.D. (1993). ZA 1296 - An Evaluation of Mutagenic Potential Using *S. typhimurium* and *E. coli*. Zeneca's Central Toxicology Laboratory (CTL), Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4206. Laboratory Study No. YV3205. December 22, 1993. Unpublished.

44373527 Griffiths, K. and Mackay, J.M. (1994). ZA 1296: An Evaluation in the Mouse Micronucleus Test. Zeneca Central Toxicology Laboratory (CTL), Alderley Park Macclesfield, Cheshire, UK. Laboratory Project ID. CTL/P/4249. February 11, 1994. Unpublished.

44505015 Robinson, P. (1996) 2-Nitro-4-methylsulfonyl benzoic acid: acute oral toxicity to the rat. Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P5210, Study No. AR6309. October 4, 1996. Unpublished.

44505016 Lees, D. (1996) AMBA (2-amino-4-methylsulfonyl benzoic acid): acute oral toxicity to the rat. Central Toxicology Laboratory, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/5282, Study No. AR6339. December 11, 1996. Unpublished.

44505017 Horner, S.A. (1997) ZA1296: Acute Neurotoxicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Project Identification Number AR6196. October 30, 1997. Unpublished.

44505018 Horner, S.A. (1997) Trimethyltin chloride: Acute Neurotoxicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Project Identification Number AR6406. October 28, 1997. Unpublished.

44505019 Horner, J. M. (1995) ZA1296: 90 Day Dietary Toxicity Study in Rats. Zeneca Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/P/4456. February 8, 1995. Unpublished.

44505020 Pinto, P.J. (1997) ZA1296: 90 Day Dietary Toxicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/P/4986, November 5, 1997. Unpublished.

44505021 Brammer, A. (1995) ZA1296: 90 Day Dietary Study in Rats to Investigate Selective Non-Ocular Toxicity End Points. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/T/2887; Study No: PR0990, August 1, 1995. Unpublished.

44505022 Pinto, P.J. (1997) ZA 1296: 90 Day Dietary Toxicity Study in Mice. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire. Laboratory Project Report No. CTL/P/5488. Laboratory Project Study No. PM1062. November 5, 1997. Unpublished.

44505023 Brammer, A. (1997) ZA1296: 90 Day Oral Toxicity Study in Dogs. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/P/4945, October 1, 1997. Unpublished.

44505024 Lees, D. (1997) ZA1296: 21 Day Dermal Toxicity Study in Rabbits. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Study Number LB0575. Report #CTL/P/5035. October 23, 1997. Unpublished.

44505025 Horner, S.A. (1997) ZA1296: Subchronic Neurotoxicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Project Identification Number PR1043. October 31, 1997. Unpublished.

44505026 Pinto, P.J. (1997) ZA 1296: One Year Dietary Toxicity Study in Mice. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Project Report No. CTL/P/5682. Laboratory Project Study No. PM1062. November 12, 1997. Unpublished.

44505027 Brammer, A. (1997) ZA1296: One Year Oral Toxicity Study in Dogs. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/P/5511, November 6, 1997. Unpublished.

44505028 Rattray, N.J. (1997) ZA1296: 80 Week Carcinogenicity Study in Mice. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/5281, November 5, 1997. Unpublished.

44505029 Odum, J. (1997) ZA1296: Biochemical Studies in Rat and Mouse Liver. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No.: CTL/R/1339, November 14, 1997. Unpublished.

44505032 Moxon, M.E. (1997). ZA1296: Dose Range Finding Study in the Pregnant Rabbit. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report # CTL/L/8024, Laboratory Study # RB0677, December 11, 1997. Unpublished.

44505033 Milburn, G.M. (1997) ZA1296: Multigeneration Study in the Rat. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Study No. RR0691. Report No. CTL/P/5147. December 10, 1997. Unpublished.

44505034 Moxon, M.E. (1997) ZA1296: Multigeneration Study in the Mouse. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Study Nos. RM0728/F0, RM0728/F1, and RM0728/F2. December 4, 1997. Unpublished.

44505035 Brammer, A. (1997) ZA1296: 2 Year Dietary Toxicity and Oncogenicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/P/5481, December 16, 1997. Unpublished.

44505036 Scott, R., (1998) Pathology Characteristics of the Alderley Park Strain (AP:Alpk:SD) of Rat. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/R/1349, January 9, 1998. Unpublished.

44505037 Callander, R.D. (1996). MNBA: An Evaluation of Mutagenic Potential Using *S. typhimurium* and *E. coli*. Zeneca's Central Toxicology Laboratory (CTL), Alderly Park, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/4955. Laboratory Study No. YV3770. April 22, 1996. Unpublished.

44505038 Callander, R.D. (1996). AMBA (2-Amino-4-Methylsulfonyl Benzoic Acid): An Evaluation of Mutagenic Potential Using *Salmonella typhimurium* and *E. coli*. Zeneca's Central Toxicology Laboratory (CTL), Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report No. CTL/P/5226. Laboratory Study No. YV3941. November 28, 1996. Unpublished.

44505101 Prescott, E., Bennett, D. (1995) ZA1296: Whole body autoradiography study in the rat following a single oral dose (mg/kg). Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/4666/PR0990, September 19, 1995. Unpublished.

44505102 Macpherson, D. (1996) ZA1296: Excretion and tissue retention of a single oral dose (100 mg/kg) in the rat. Central Toxicology Laboratory, Cheshire. UK. Laboratory Report/Study No. CTL/P/4927/UR0501, May 17, 1996. Unpublished.

44505103 Gledhill, A. (1996) ZA1296: Biotransformation in the rat. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/4930/UR0442, June 3, 1996. Unpublished.

44505104 Macpherson, D. (1996) ZA1296: Excretion and tissue retention of a single oral dose (1 mg/kg) in the rat. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/4948/UR0502, May 20, 1996. Unpublished.

44505105 Macpherson, D. (1996) ZA1296: Excretion and tissue retention of a single intravenous dose (1 mg/kg) in the rat. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/4976/UR0522, May 29, 1996. Unpublished.

44505106. Macpherson, D. (1996) ZA1296: Excretion and tissue retention of a single oral dose (1 mg/kg) in the rat following repeat dosing. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/4995/UR0525, May 24, 1996. Unpublished.

44505110 Provan, W.M. Whitfield, A.C., Naylor, J.L. (1994). The Extent of Tyrosinemia and Ocular Lesions Induced by 50 Triketone Herbicides in Rats. Zeneca Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No. CTL/R/1204, October 30, 1994. Unpublished.

44505111 Milburn, G. M., (1997) ZA1296: 28 Day Toxicity Study in the Rat. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1308; Study No: XR6167, November 18, 1997. Unpublished.

44505112 Williams, J., (1997) ZA1296: A Single Generation Reproductive Toxicity Study in the Rat (up to day 5 *post partum*). Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/L/8015; Study No: XR6312/F0. November 11, 1997. Unpublished.

44505113 Hall, M.G., (1996) ZA1296: Systemic Exposure Following Dietary Administration. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/L/7179, June 10, 1996. Unpublished.

44505114 Hall, M.G. (1997) ZA1296: Preliminary Data from a Single Oral Dosing Study in Man. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No.: CTL/R/1352. Study No.: QH0019, December 18, 1997. Unpublished.

44505115 Stevens, V. (1998) A Single Dose, Crossover, Pharmacokinetic Study of Two Formulations of NTBC (SC0735) in Healthy Volunteers. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No.: CTL/R/1360; /Study No.: CCT96/001, January 9, 1998. Unpublished.

44505116 Brammer, A., Provan, W.M. (1997) ZA1296: 90 Day Dietary Dose Response Study in Mice. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1316; Study No: XR6168, December 19, 1997. Unpublished.

44537101 Gledhill, A.J. (1997) ZA1296: Excretion, tissue distribution and metabolism of a single oral dose (1 mg/kg and 100 mg/kg) in the mouse. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/R/1331/UM0573, November 13, 1997. Unpublished.

44537103 Brammer, A., Provan, W.M. (1997) ZA1296: 90 Day Reversibility Studies in Rats. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1302; Study Nos: PR1066 and XR6242, December 22, 1997. Unpublished.

44537104 Tinston, D.J. (1997) ZA1296: Ocular toxicity Development and reversibility Study in Rats. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report/Study No. CTL/P/5001/PR 1022, April 23, 1997. Unpublished.

44537105 Robinson, M., (1995) A Study to Investigate the Morphology and Pathology of Ocular Lesions in Alpk:AP$_f$SD Rats Fed Different Levels of Tyrosine in a Low Protein Diet. Central Toxicology Laboratory, Cheshire, UK. Laboratory Report No.: CTL/R/1243, June 21, 1995. Unpublished.

44537106 Brammer, A., (1997) ZA1296: 90 Day Dose Response Study in Male Rats. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1304; Study No: XR5997, November 19, 1997. Unpublished.

44537107 Brammer, A., (1997) ZA1296: 90 Day Dose Response Study in Female Rats. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1315; Study No: XR6195, November 19, 1997. Unpublished.

44901706 Milburn, G. M., (1998) MNBA: 28 Day Oral Toxicity Study in Rats. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/5578; Study No: KR1281, January 12, 1998. Unpublished.

44901707 Moxon, M.E. (1999). First Revision to ZA1296: Developmental Toxicity Study in the Rabbit. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report # CTL/P/4892, Laboratory Study # RB0684, June 28, 1999. Unpublished.

44901708 Diggins, G. (1997). ZA1296: Preliminary Developmental Toxicity Study in the Mouse. Quintiles Toxicology/Pathology Services, Quintiles England Limited, Ledbury, Herefordshire, UK. Laboratory Project ID ICL/022/97, September 5, 1997. Unpublished.

44901711 Elcombe, B. M., Meadowcroft, S., (1998) ZA1296: Effects of AMBA, a Metabolite of ZA1296 on p-Hydroxyphenylpyruvate Dioxygenase (HPPD) Activity. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1361; Study No: XR6243, April 1998. Unpublished.

44901712 Elcombe, B. M., Meadowcroft, S., (1998) ZA1296: Effects of MNBA, a Metabolite of ZA1296 on p-Hydroxyphenylpyruvate Dioxygenase (HPPD) Activity. Central Toxicology Laboratory, Cheshire, UK, Laboratory Report No: CTL/R/1367; Study No: XR6244., April 1998. Unpublished.

44920801 Moxon, M.E. (1999). First Revision to ZA1296: Developmental Toxicity Study in Rats. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report # CTL/P/5034, Laboratory Study # RR0700, June 28, 1999. Unpublished.

44920802 Moxon, M.E. (1999). ZA1296: Developmental Toxicity Study in Mice. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Laboratory Report # CTL/P/6238, Laboratory Study # RM0793, June 29, 1999. Unpublished.

44920803 Hall, M.G. (1998) ZA1296: Investigation of Systemic Exposure Following a Single Dermal Application of Spray Formulations to Healthy Male Volunteers. Central Toxicology Laboratory, Cheshire, UK. Laboratory Project Identification Number QH0020. July 24, 1998. Unpublished.

45196004 Lees, D. (1996) MNBA: Acute dermal toxicity study in rats. Central Toxicology Laboratory, Alderley Park, Macclesfield, Cheshire, UK. Report ID CTL/P/4963, May 12, 1996. Unpublished.

9.0 APPENDICES

Tables for Use in Risk Assessment

9.1 Risk Assessment Toxicity Profile Summary Tables

9.1.1 Acute Toxicity Table - See Section 4.1

9.1.2 Subchronic, Chronic, and Other Toxicity Table

The summary of toxicological dose and endpoints for mesotrione for use in human risk assessment is presented below[1]:

Exposure Scenario	Dose Used in Risk Assessment, UF	FQPA SF and LOC for Risk Assessment	Study and Toxicological Effects
Acute Dietary all populations	Not Applicable	Not Applicable	No appropriate study available
Chronic Dietary all populations	LOAEL= 2.1 mg/kg/day UF =3 **Chronic RfD** = 0.007 mg/kg/day	FQPA SF = 10X **cPAD** = chronic RfD / FQPA SF = 0.0007 mg/kg/day	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups
Short-Term Incidental Oral (1-7 days) (Residential)	NOAEL = 100 mg/kg/day	**LOC for MOE** = 1000 (Residential)	Developmental Toxicity Study - rat Maternal NOAEL = 100 mg/kg/day based upon decreased body weight gains during treatment and decreased food consumption
Intermediate-Term Incidental Oral (7 days - several months) (Residential)	LOAEL = 2.1 mg/kg/day	**LOC for MOE** = 3000 (Residential)	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups
Short-Term Dermal (1-7 days) (Occupational/ Residential)	Oral study LOAEL = 100 mg/kg/day (dermal absorption rate = 25%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Developmental Toxicity Study - rat Developmental LOAEL = 100 mg/kg/day based upon delays in skeletal ossification and changes in *manus/pes* ossification assessments
Intermediate-Term Dermal (1 week - several months) (Occupational/ Residential)	Oral study LOAEL = 2.1 mg/kg/day (dermal absorption rate = 25%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups

(Table continues, footnotes follow)

Exposure Scenario	Dose Used in Risk Assessment, UF	FQPA SF and LOC for Risk Assessment	Study and Toxicological Effects
Long-Term Dermal (several months - lifetime) (Occupational/ Residential)	Oral study LOAEL = 2.1 mg/kg/day (dermal absorption rate = 25%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups
Short-Term Inhalation (1-7 days) (Occupational/ Residential)	Oral study LOAEL = 100 mg/kg/day (inhalation absorption rate = 100%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Developmental Toxicity Study - rat Developmental LOAEL = 100 mg/kg/day based upon delays in skeletal ossification and changes in *manus/pes* ossification assessments
Intermediate-Term Inhalation (1 week - several months) (Occupational/ Residential)	Oral study LOAEL = 2.1 mg/kg/day (inhalation absorption rate = 100%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups
Long-Term Inhalation (several months - lifetime) (Occupational/ Residential)	Oral study LOAEL = 2.1 mg/kg/day (inhalation absorption rate = 100%)	**LOC for MOE** = 300 (Occupational) **LOC for MOE** = 3000 (Residential)	Reproduction Study - mouse Offspring LOAEL = 2.1 mg/kg/day based upon tyrosinemia in F_1 and F_{2a} offspring and ocular discharge in F_1 pups
Cancer (oral, dermal, inhalation)	"not likely"	not applicable	Acceptable oral rat and mouse carcinogenicity studies; no evidence of carcinogenic or mutagenic potential.

[1] UF = uncertainty factor, FQPA SF = FQPA safety factor, NOAEL = no observed adverse effect level, LOAEL = lowest observed adverse effect level, PAD = population adjusted dose (a = acute, c = chronic) RfD = reference dose, MOE = margin of exposure, LOC = level of concern

9.2 Toxicity Profile of Mesotrione Technical and its Metabolites

9.2.1 Toxicity Profile of Mesotrione Technical (Major Studies)

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.3100 90-Day oral toxicity rodents (rat)	44505020 (1997) Acceptable/guideline 0, 2.5, 5, 7.5, or 150 ppm M: 0, 0.21, 0.41, 0.63, or 12.46 mg/kg/day F: 0, 0.23, 0.47, 0.71, or 14.48 mg/kg/day	NOAEL = 0.41/0.47 mg/kg/day (M/F) LOAEL = 0.63/0.71 mg/kg/day (M/F), based upon corneal lesion in males
870.3100 90-Day oral toxicity rodents (rat)	44505019 (1995) Acceptable/guideline 0, 1, 125, 1250, or 12500 ppm M: 0, 0.09, 11, 112, or 1111 mg/kg/day F: 0, 0.10, 13, 126, or 1213 mg/kg/day	NOAEL = 0.09/0.10 mg/kg/day (M/F) LOAEL = 11/13 mg/kg/day (M/F), based upon corneal abnormalities in both sexes and decreased body weight gain in males
870.3100 90-Day oral toxicity rodents (mouse)	44505022 (1997) Acceptable/guideline 0, 10, 50, 350, or 7000 ppm M: 0, 1.7, 8.4, 61.5, or 1212 mg/kg/day F: 0, 2.4, 12.4, 80.1, or 1537 mg/kg/day	NOAEL = 1212/1537 mg/kg/day (M/F) LOAEL > 1212/1537 mg/kg/day (M/F) No effects noted
870.3150 90-Day oral toxicity in nonrodents (dog)	44505023 (1997) Acceptable/guideline M & F: 0, 100, 600, or 1000 mg/kg/day	NOAEL = 1000 mg/kg/day (M/F) LOAEL > 1000 mg/kg/day (M/F) No effects noted
870.3200 21/28-Day dermal toxicity (rabbit)	44505024 (1997) Acceptable/guideline M & F: 0, 10, 500 or 1000 mg/kg/day	NOAEL = 1000 mg/kg/day (M/F) LOAEL > 1000 mg/kg/day (M/F) No systemic effects noted
870.3250 90-Day dermal toxicity	NA	NA
870.3465 90-Day inhalation toxicity	NA	NA

table continues next page 66

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.3700a Prenatal developmental in rodents (rat)	44920801 (1999) Acceptable/guideline F: 0, 100, 300, or 1000 mg/kg/day	**Maternal** NOAEL = 100 mg/kg/day LOAEL = 300 mg/kg/day, based upon decreased maternal body weight gains during treatment and decreased food consumption **Developmental** NOAEL not established LOAEL = 100 mg/kg/day, based upon delays in skeletal ossification and changes in *manus/pes* ossification assessments
870.3700a Prenatal developmental in rodents (mouse)	44920802 (1999), 44901708 (1997) Acceptable F: 0, 10, 60, 150, or 600 mg/kg/day	**Maternal** NOAEL ≥600 mg/kg/day LOAEL was not observed **Developmental** NOAEL = 150 mg/kg/day LOAEL = 600 mg/kg/day, based upon decreased ossification of the cervical vertebrae centra
870.3700b Prenatal developmental in nonrodents (rabbit)	44901707 (1997), 44505032 (1999) Unacceptable/not upgradable F: 0, 100, 250, or 500 mg/kg/day	**Maternal** NOAEL = 100 mg/kg/day LOAEL = 250 mg/kg/day based upon abortions and clinical signs of toxicity **Developmental** NOAEL not established LOAEL = 100 mg/kg/day based upon delayed ossification of the 7th cervical transverse process and odontoid and increases in extra full 13th ribs and 27 pre-sacral vertebra
870.3800 Reproduction and fertility effects (rat)	44505033 (1997) Acceptable/guideline 0, 2.5, 10, 100 or 2500 ppm M: 0, 0.3, 1.1, 11.7, or 287.7 mg/kg/day F: 0, 0.3, 1.2, 12.4, or 311.4 mg/kg/day	**Parental/Systemic** NOAEL not established LOAEL = 0.3 mg/kg/day (M/F), based upon significantly increased plasma tyrosine levels and increased liver wts. in F_2 males **Offspring/Systemic** NOAEL not established LOAEL = 0.3 mg/kg/day (M/F), based upon significantly increased plasma tyrosine levels in F_2 male pups **Reproductive** NOAEL = 0.3 mg/kg/day LOAEL = 1.1/1.2 mg/kg/day (M/F), based upon decreased F_2 mean litter size
870.3800 Reproduction and fertility effects (mouse)	44505034 (1997) Acceptable/guideline 0, 10, 50, 350, 1500 or 7000 ppm M: 0, 2.1, 10.1, 71.4, 306.7 or 1455.5 mg/kg/day F: 0, 2.4, 11.7, 82.5, 362.7 or 1652.3 mg/kg/day	**Parental/Systemic** NOAEL = 10.1/11.7 (M/F) LOAEL = 71.4/82.5 mg/kg/day (M/F), based upon increased kidney weights and tyrosinemia in the and F_1 males **Offspring/Systemic** NOAEL not established LOAEL = 2.1/2.4 mg/kg/day (M/F), based upon tyrosinemia and ocular discharge in the F_1 and F_2 offspring **Reproductive** NOAEL = 1455.5/1652.3 mg/kg/day (M/F) LOAEL > 1455.5/1652.3

table continues next page 67

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.4300 Combined chronic toxicity/ carcinogenicity rodents (rat)	44505035 (1997), 44505036 (1998) Acceptable/guideline 0, [1.0, 2.5], 7.5, 100 or 2500 ppm M: 0, [0.06, 0.16], 0.48, 6.48 or 159.9 mg/kg/day F: 0, [0.08, 0.19], 0.57, 7.68, or 189.5 mg/kg/day	NOAEL = 0.16/0.19 mg/kg/day (M/F) [The NOAEL only applies to ocular lesions; a NOAEL was not determined for kidney and liver weights or hepatocyte fat vacuolation in males] LOAEL = 0.48/0.57 mg/kg/day (M/F), based upon ocular lesions, increases in kidney and liver weights, and hepatocyte fat vacuolation in males **No evidence of carcinogenicity**
870.4100a Chronic toxicity rodents (mouse)	44505026 (1997) Acceptable/guideline 0, 10, 50, 350 or 7000 ppm M: 0, 1.5, 7.8, 56.2 or 1114 mg/kg/day F: 0, 2.1, 10.3, 72.4 or 1494.5 mg/kg/day	NOAEL = 56.2/72.4 mg/kg/day (M/F) LOAEL = 1114/1494.5 mg/kg/day (M/F), based upon decreases in body weight gain and food utilization in males
870.4100b Chronic toxicity nonrodents (dog)	44505027 (1997) Acceptable/guideline M & F: 0, 10, 100 or 600 mg/kg/day	NOAEL was not established LOAEL = 10 mg/kg/day, based upon evidence of tyrosinemia in both sexes and increased incidence of erythrophagocytosis in the mesenteric lymph nodes of females
870.4200b Carcinogenicity mouse	44505028 (1997) Acceptable 0, 10, 350 or 7000 ppm M: 0, 1.4, 49.7 or 898 mg/kg/day F: 0, 1.8, 63.5 or 1103 mg/kg/day	NOAEL = 49.7/63.5 mg/kg/day (M/F) LOAEL = 898/1103 mg/kg/day (M/F), based upon decreased body weight, body weight gain, and food efficiency in males **no evidence of carcinogenicity**
Gene Mutation 870.5100 reverse gene mutation assay in bacteria	44373526 (1993) Acceptable	There was *no evidence* of induced mutant colonies over background
Gene Mutation 870.5300 *in vitro* forward gene mutation assay in mouse lymphoma cells	44373525 (1994) Acceptable	There were no treatment-related increases in mutant frequency in the presence or absence of S9 activation
Cytogenetics 870.5375 *in vitro* mammalian cytogenetics assay	44353724 (1994) Acceptable	Not clastogenic with S9 activation and equivocal for clastogenic activity without S9 activation

table continues next page

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.5395 bone marrow micronucleus assay	44373527 (1994) Acceptable	There was no significant increase in the frequency of micronucleated polychromatic erythrocytes in bone marrow after any treatment time.
870.6200a Acute neurotoxicity screening battery	44505017 (1997), 44505018 (1997) Acceptable M & F: 0, 20, 200 or 2000 mg/kg/day	NOAEL = 2000 mg/kg/day (M/F) LOAEL > 2000 mg/kg/day (M/F) No effects noted
870.6200a Subchronic neurotoxicity screening battery	44505025 (1997) Acceptable/guideline M: 0, 0.2, 8.25 or 403 mg/kg/day F: 0, 0.23, 9.29 or 467 mg/kg/day	NOAEL = 0.20/0.23 mg/kd/day (M/F) LOAEL = 8.25/9.29 mg/kg/day (M/F), based upon corneal opacities and/or vascularization of the cornea of the eye in males
870.6300 Developmental neurotoxicity	NA	NA
870.7485 Metabolism (rat)	44505101 (1995), 44505102 (1996), 44505103 (1996), 44505104 (1996), 44505105 (1996), 44505106 (1996) Acceptable M & F: 1 or 100 mg/kg single oral dose, 1 mg/kg single iv dose, repeated dose at 1 mg/kg; special study - 50 mg/kg single oral dose	Mesotrione was readily absorbed and distributed in the body. Tissue distribution was about the same in both sexes, although one study showed higher residues in the kidneys in females, with the highest residues of the test compound in the liver and kidney. Higher doses resulted in higher residues in the liver and kidney, while repeated doses resulted in reduced accumulation of residues in all tissues. Levels of radioactivity in tissues of iv-dosed animals were essentially the same as in orally-dosed animals. Over 50% of the administered dose was excreted in the urine in both sexes and around 25% was excreted in the feces within 72 hours. Females exhibited slightly higher total urinary excretion than males, but total fecal excretion was about the same in both sexes. Increasing the dose or repeated doses had little effect on the pattern of excretion in both sexes. The overall pattern of excretion was similar between orally-dosed and iv-dosed rats. The metabolite profile was similar between the sexes in each group and between the single-dosed and repeated-dosed animals. The parent compound, mesotrione, was the major component identified in the urine (> 47%) and feces (> 55%). Minor metabolites identified were MNBA, AMBA, 5-hydroxymesotrione, and 4-hydroxymesotrione.

table continues next page

Guideline No./ Study Type	MRID No. (year)/ Classification /Doses	Results
870.7485 Metabolism (mouse)	44537101 (1997) M & F: 1 or 100 mg/kg single oral dose	Metabolism in the mouse was very similar to the rat (above) except that males had slightly increased total fecal excretion when compared to females and, females in the low-dose group excreted higher (1.5x) levels of parent compound in the urine than males. Free mesotrione was the major component in the urine and feces (≥ 50% of the dose). Minor components in the fecal extracts included AMBA and MNBA.
870.7600 Dermal penetration	NA	NA

9.2.2 Toxicity Profile of Mesotrione Technical (Other Studies)

Study Type	MRID No. (year)/ /Doses	Results
Non-guideline reversibility (rat)	44537103 (1997) 0, 5, 100, or 2500 ppm 0, 0.37, 7.52, and 192 mg/kg/day (males)	Tyrosinemia with associated increases in ocular lesions, changes in liver enzyme activities, and adaptive changes in liver and kidney weights. Upon removal of mesotrione from the diet, complete recovery was observed in all parameters except for only partial recovery of HPPD activity and liver weights
Non-guideline Ocular toxicity development and reversibility study (rat)	44537104 (1997) 2500 ppm 272 mg/kg/day (males)	Corneal lesions were resolved ophthalmoscopically and histologically following an 8 week recovery period.
Non-guideline Ocular lesions in rats fed tyrosine	44537105 (1995) Low protein diet supplemented with 0, 0.5, 1.0, 2.5, or 5% (w/w) L-tyrosine	2.5 and 5% tyrosine induced ocular toxicity
Non-guideline 90-day dose-response study (rat)	44537106 (1997) 0, 0.5, 1, 3, 4, 5, 7.5, 10, or 100 ppm 0, 0.04, 0.09, 0.27, 0.35, 0.44, 0.67, 0.89, and 8.96 mg/kg/day (males)	Tyrosinemia with associated increases in ocular lesions and urinary phenolic acids output, changes in liver enzyme activities, and adaptive changes in liver and kidney weights
Non-guideline 90-day dose-response study (rat)	44537107 (1997) 0, 1, 5, 10, 50, 100, 1000, or 2500 ppm 0, 0.09, 0.48, 0.95, 4.82, 9.54, 94.83, and 236.75 mg/kg/day (females)	Tyrosinemia with associated increases in ocular lesions and urinary phenolic acids output, changes in liver enzyme activities, and adaptive changes in liver weights
Non-Guideline Single Generation Reproductive Study (rat)	44505112 (1997) Mesotrione (0 or 2500 ppm) and/or L-tyrosine (0, 0.5, 1, or 2%)	Decreased litter size and pup viability following treatment of the maternal animals with 2500 ppm mesotrione + 0.5% or 1% tyrosine; males more susceptible
Non-guideline Effect on tyrosinemia in female rats given a high-tyrosine diet	44505111 (1997) 0, 0.5, 1.0, or 2.5% tyrosine in diet or 100 ppm mesotrione diet with 0, 0.5, 1.0, or 2.5% tyrosine	Mesotrione + tyrosine in the diet caused marked tyrosinemia and associated ocular lesions and changes in liver enzyme activities. Treatment with a combination of mesotrione and tyrosine caused more marked effects than treatment with either compound alone.
Non-guideline 7-day study (rat)	44505113 (1996) Two batches at 1, 40, 25, 1250, or 5000 ppm (males)	Systemic exposure comparisons based on urinary excretion of mesotrione and its effect on plasma tyrosine concentrations showed no significant differences between Batch P8 and P11

table continues next page

Study Type	MRID No. (year)/ /Doses	Results
Non-guideline 90-day dose-response study (mouse)	44505116 (1997) 0, 1, 10, 50, 100, 350, 1000, 3500, or 7000 ppm M:0, 0.16, 1.69, 8.49, 17.95, 58.46, 179.27, 599.85, or 1222.53 mg/kg/day F: 0,0.19, 1.94, 10.80, 20.46, 72.70, 214.88, 714.76, or 1436.40 mg/kg/day	Tyrosinemia with associated increases in urinary phenolic acids output and changes in liver enzyme activities
Non-guideline 90-day dose-response study (rat)	44505021 (1995) 0, 10, 20, 50, or 125 ppm 0, 0.9, 1.7, 4.3, or 10.7 mg/kg/day (males)	Dose-dependent changes in corneal opacity and corneal vascularization. Non-dose-dependent changes in adjusted (to body) kidney and liver weights
Non-Guideline; Biochemical Studies in Rat and Mouse Liver	44505029 (1997) 0, 1000, 3000 (mice only) 7000, or 16000 (rats only) ppm	Slight increases in rat liver weights, slight microscopic liver hypertrophy in rats and mice, and minimal CYP induction in both species

72

9.2.3 Toxicity Profile of MNBA and AMBA (Mesotrione Metabolites)

Study Type	MRID No.(year)/ Classification/Doses	Results
MNBA		
Gene Mutation 870.5100 Reverse gene mutation assay in bacteria	44505037 (1996) Unacceptable/not upgradable 100 to 5000 μg/plate ± S9	There was *no evidence* of induced mutant colonies over background except for an equivocal response with tester strain TA100 at 5000 μg/plate (+S9).
Non-Guideline 28-Day Oral Toxicity Study	44901706 (1998) 0, 15, 150, or 1000 (limit dose) mg/kg/day, males and females	Increase in motor activity in females
Non-Guideline Effects on HPPD activity	44901712 (1998) 0.02 or 20 μM in isolated male liver cytosol	Weak inhibition of HPPD *in vitro*
AMBA		
Gene Mutation 870.5100 Reverse gene mutation assay in bacteria	44505038 (1996) Acceptable 100 to 5000 μg/plate ± S9	There was *no evidence* of induced mutant colonies over background
Non-Guideline Effects on HPPD activity	44901711 (1998) 0.02 or 20 μM in isolated male liver cytosol	Weak inhibition of HPPD *in vitro*

9.2.4 Toxicity Profile of Mesotrione and NTBC (Mesotrione Analog) in Humans

Study Type	MRID No.(year)/ Classification/Doses	Results
Mesotrione		
Non-Guideline Preliminary Data from a Single Oral Dosing Study	44505114 (1997) Unacceptable (non-guideline) 0.1, 0.5, or 4 mg/kg (males)	Elevated plasma tyrosine levels
Non-Guideline Single Dermal Application	44920803 (1998) Acceptable (non-guideline) 4 or 25.6 mg (males)	Remained in the stratum corneum and had no effect on plasma tyrosine concentration
NTBC		
Non-Guideline Single Dose Study	44505115 (1998) Acceptable (non-guideline) 1 mg/kg (males) in either liquid or capsule	Linear increase in mean plasma tyrosine levels until 48 hours after dosing and dosing formulation had no effect on bioavailability

74

10.0 ATTACHMENTS

The following attachments contain the DERs and Study Overviews for mesotrione:

MRID No. 44373512. Acute Oral Toxicity. (Electronic copy available: Filename: 44373512.der)

MRID No. 44373514. Acute Dermal Toxicity. (Electronic copy available: Filename: 44373514.der)

MRID No. 44373516. Acute Inhalation Toxicity. (Electronic copy available: Filename: 44373516.der)

MRID No. 44373518. Primary Eye Irritation Study. (Electronic copy available: Filename: 44373518.der)

MRID No. 44373520. Primary Dermal Irritation Study. (Electronic copy available: Filename: 44373520.der)

MRID No. 44373522. Dermal Sensitization Study. (Electronic copy available: Filename: 44373522.der)

MRID No. 44373526. *Salmonella typhimurium/E. coli*. mammalian activation gene mutation assay. (Electronic copy available: Filename: 44373526.der)

MRID No. 44373525. Mouse Lymphoma Cell Forward Mutation Assay. (Electronic copy available: Filename: 44373525.der)

MRID No. 44373524. *In vitro* Chromosome Aberration Assay in Human Lymphocytes. (Electronic copy available: Filename: 44373524.der)

MRID No. 44373527. *In vivo* mammalian cytogenetics - micronucleus assay in mice. (Electronic copy available: Filename: 44373527.der)

MRID No. 44505019. Subchronic Oral Toxicity in Rats. (Electronic copy available: Filename: 44505019.der)

MRID No. 44505020. Subchronic Oral Toxicity in Rats. (Electronic copy available: Filename: 44505020.der)

MRID No. 44505022. Subchronic Oral Toxicity in Mice. (Electronic copy available: Filename: 44505022.der)

~~73B~~ 75

MRID No. 44505023. Subchronic Oral Toxicity in Dogs. (Electronic copy available: Filename: 44505023.der)

MRID Nos. 44505035, 44505036. Combined Chronic/Carcinogenicity Study in Rats. (Electronic copy available: Filename: 44505035.der)

MRID No. 44505028. Carcinogenicity Study in Mice. (Electronic copy available: Filename: 44505028.der)

MRID No. 44505026. Chronic Oral Toxicity Study in Mice. (Electronic copy available: Filename: 00101638.der)

MRID No. 44505027. Chronic Toxicity in Dogs. (Electronic copy available: Filename: 44505027.der)

MRID No. 44920801. Prenatal Developmental Toxicity Study in Rats. (Electronic copy available: Filename: 44950801.der)

MRID Nos. 44901707, 44505032. Prenatal Developmental Toxicity Study in Rabbits. (Electronic copy available: Filename: 44901707.der)

MRID Nos. 44920802, 44901708. Prenatal Developmental Toxicity Study in Mice. (Electronic copy available: Filename: 44920802.der)

MRID No. 44505033. Multigeneration Reproduction Toxicity Study in Rats. (Electronic copy available: Filename: 44505033.der)

MRID No. 44505034. Multigeneration Reproduction Toxicity Study in Mice. (Electronic copy available: Filename: 44505034.der)

MRID Nos. 44505017, 44505018. Acute Neurotoxicity in Rats. (Electronic copy available: Filename: 44505017.der)

MRID No. 44505025. 90-Day Neurotoxicity in Rats. (Electronic copy available: Filename: 44505025.der)

MRID No. 44505024. 21-Day Dermal Toxicity Study in Rats. (Electronic copy available: Filename: 44693801.der)

MRID Nos. 44505101 through 44505106. Metabolism Study in Rats. (Electronic copy available: Filename: 44505101.der)

73e
76

MRID No. 44537101. Metabolism Study in Mice. (Electronic copy available: Filename: 44537101.der)

MRID No. 44537104. Ocular Toxicity Study in Rats. (Electronic copy available: Filename: 44537104.der)

MRID No. 44505114. Single Oral Dosing Study in Man.. (Electronic copy available: Filename: 44505114.der)

MRID No. 44505115. Pharmacokinetic Study of Two Formulations of NTBC (Mesotrione Analogue) in Humans. (Electronic copy available: Filename: 44505115.der)

MRID No. 44920803. Single-Dose Dermal Application Study in Humans. (Electronic copy available: Filename: 44920803.der)

MRID No. 44505112. Single Generation Reproductive Study in Rats. (Electronic copy available: Filename: 44505112.der)

MRID No. 44505111. Effect on Tyrosinemia in Female Rats on a High Tyrosine Diet. (Electronic copy available: Filename: 44505111.der)

MRID No. 44537107. 90-Day Dose Response Study in Female Rats. (Electronic copy available: Filename: 44537107.der)

MRID No. 44537106. 90-Day Dose Response Study in Male Rats. (Electronic copy available: Filename: 44537106.der)

MRID No. 44537105. Ocular Lesions in Rats Fed Tyrosine. (Electronic copy available: Filename: 44537105.der)

MRID No. 44505029. Biochemical Studies in Rat and Mouse Liver. (Electronic copy available: Filename: 44505029.der)

MRID No. 44505113. 7-Day Oral Toxicity Study in Rats. (Electronic copy available: Filename: 44505113.der)

MRID No. 44537103. 90-Day Reversibility Study in Male Rats. (Electronic copy available: Filename: 44537103.der)

MRID No. 44505021. 90-Day Dietary Special Study in Rats. (Electronic copy available: Filename: 44505021.der)

735
77

MRID No. 44505116. 90-Day Dose Response Study in Mice. (Electronic copy available: Filename: 44505116.der)

MRID No. 44505110. Extent of Tyrosinemia and Ocular Lesions Induced by 50 Triketone Herbicides in Rats. (Electronic copy available: Filename: 44505110.der)

Formulation (480 g/L SC)

MRID No. 44373513. Acute Oral Toxicity. (Electronic copy available: Filename: 44373513.der)

MRID No. 44373515. Acute Dermal Toxicity. (Electronic copy available: Filename: 44373515.der)

MRID No. 44373517. Acute Inhalation Toxicity. (Electronic copy available: Filename: 44373517.der)

MRID No. 44373519: Primary Eye Irritation Study. (Electronic copy available: Filename: 44373519.der)

MRID No. 44373521. Primary Dermal Irritation Study. (Electronic copy available: Filename: 44373521.der)

MRID No. 44373523. Dermal Sensitization Study. (Electronic copy available: Filename: 44373523.der)

Metabolites

MRID No. 44505016. Acute Oral Toxicity (AMBA). (Electronic copy available: Filename: 44505016.der)

MRID No. 44505015. Acute Oral Toxicity (MNBA). (Electronic copy available: Filename: 44505015.der)

MRID No. 45196004. Acute Dermal Toxicity - Rat (MNBA). (Electronic copy available: Filename: 45196004.der)

MRID No. 44505038. *Salmonella typhimurium/E. coli.* mammalian activation gene mutation assay (AMBA). (Electronic copy available: Filename: 44505038.der)

MRID No. 44505037. *Salmonella typhimurium/E. coli.* mammalian activation gene mutation assay (MNBA). (Electronic copy available: Filename: 44505037.der)

MRID No. 44901706. 28-Day Oral Toxicity Study in Rats (MNBA). (Electronic copy available: Filename: 44901706.der)

MRID No. 44901711. Effects of AMBA on HPPD activity. (Electronic copy available: Filename: 44901711.der)

MRID No. 44901712. Effects of MNBA on HPPD activity. (Electronic copy available: Filename: 44901712.der)

C14649

79