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OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

TXR NO. 0051579

DATE: February 19, 2003

MEMORANDUM

SUBJECT: ***MEPIQUAT CHLORIDE*** - 2nd Report of the Hazard Identification Assessment Review Committee.

FROM: Brenda Tarplee, Senior Scientist
Science Information Management Branch
Health Effects Division (7509C)

THROUGH: Jess Rowland, Co-Chair
and
Elizabeth Doyle, Co-Chair
Hazard Identification Assessment Review Committee
Health Effects Division (7509C)

TO: Margarita Collantes
Registration Action Branch 2
Health Effects Division (7509C)

PC Code: 109101

On January 22, 2003 the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) reassessed FQPA requirements in response to questions posed by the Natural Resources Defense Council (NRDC). HIARC also reviewed the previous recommendation for a developmental neurotoxicity study in rats and subsequent need for a data base factor to account for this data gap. No new data have been reviewed and no changes were made to the toxicology endpoints previously selected for mepiquat chloride. This document revises the previous HIARC report dated October 21, 1999 (TXR NO. 013809).

Committee Members in Attendance

Members present were: Ayaad Assaad, William Burnam, Paula Deschamp, Elizabeth Doyle, Pamela Hurley, John Liccione, Susan Makris, Elizabeth Mendez, David Nixon, Jess Rowland, PV Shah (for Jonathan Chen), and Brenda Tarplee (Executive Secretary).

Member(s) in absentia were: Jonathan Chen

Also in attendance were: Karen Whitby (RAB1) and Ed Zager (HED IO).

Meeting materials prepared by: Brenda Tarplee, Senior Scientist, SIMB

INTRODUCTION

August 31, 1999, the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) re-evaluated the toxicology data base (including a not previously evaluated rabbit developmental toxicity study), re-established the Reference Dose (RfD) and reviewed the previously selected toxicological endpoints for acute dietary as well as occupational and residential exposure risk assessments. The HIARC also addressed the potential for enhanced sensitivity of infants and children to mepiquat chloride as required by the Food Quality Protection Act (FQPA) of 1996.

On January 22, 2003 the Health Effects Division (HED) Hazard Identification Assessment Review Committee (HIARC) reassessed FQPA requirements in response to questions posed by the Natural Resources Defense Council (NRDC). HIARC also reviewed the previous recommendation for a developmental neurotoxicity study in rats and subsequent need for a data base factor to account for this data gap. No new data have been reviewed and no changes were made to the toxicology endpoints previously selected for mepiquat chloride. This document revises the previous HIARC report dated October 21, 1999 (TXR NO. 013809).

I. FQPA HAZARD CONSIDERATIONS

1. Adequacy of the Toxicity Data Base

The HIARC concluded that the toxicology database for mepiquat chloride is not complete for FQPA assessment.

On August 31, 1999, HIARC requested that the following studies be conducted with mepiquat chloride:

- Acute neurotoxicity study - rats
- Subchronic neurotoxicity study - rats
- Developmental neurotoxicity study - rats

2. Evidence of Neurotoxicity

The HIARC concluded that there is a concern for neurotoxicity resulting from exposure to mepiquat chloride.

There are currently no neurotoxicity studies (acute or subchronic) conducted with mepiquat chloride available at this time.

3. Developmental Toxicity Study Conclusions

Executive Summary for a Developmental Toxicity Study in Rats
[Guideline No. 83-3a (870.3700), MRID 42337101]

In this developmental toxicity study, pregnant Wistar strain rats, 25/group, were administered aqueous solutions of Mepiquat chloride by gavage during gestation days 6 through 15. The doses used were 0, 50, 150 or 300 mg/kg b.w./day and were based on the results of the range finding

study in which 100, 300 or 600 mg of Mepiquat chloride/kg b.w./day were tested. The purity (active ingredient content) of Mepiquat chloride was 57.9%.

Treatment-related **maternal effects** were observed only in the high-dose (300 mg/kg) group and included clinical signs of toxicity and decreases in the food consumption and body weight gain. These effects were not observed when dosing with Mepiquat chloride was discontinued. There were no unscheduled mortalities.

Most dams (22-24/25) in the high-dose group showed pronounced but reversible tremors, unsteady gait, indrawn flanks and hypersensitivity; whereas 4/25 dams also had ataxia. All of these findings were noted at approximately 1.5-2.0 hours after dosing, lasted for about 4 hours and, with the exception of ataxia, were less frequent during the second half of the treatment period. Ataxia was observed in 2 dams during gestation day (g.d.) 7 only, in one dam during g.d. 8 and in another dam during g.d. 9.

Compared with the control values, food consumption of the high-dose dams was reduced by 10.4 to 19.1% ($p<0.01$) during the greater part of the dosing period (g.d. 6-13), but not thereafter. Mean body weight gains were also reduced during the same period by 16.2-65.1% ($p<0.01$), when compared with the control values. However, when mean body weights on g.d. 20 were corrected for uterine weights, the high-dose dams weighed only 13% less than did the controls and this difference was statistically insignificant.

Mepiquat chloride, at the three levels tested, had no effect on any of the **developmental toxicity** parameters examined. No embryotoxicity, fetotoxicity and no indications of any teratogenic effects were observed in this study.

Based on the clinical signs of toxicity and decreases in the food consumption and body weight gains, the **Maternal Toxicity LOAEL is 300 mg/kg/day and the Maternal Toxicity NOAEL is 150 mg/kg/day.**

Since developmental toxicity was not observed in this study, the **Developmental Toxicity NOAEL is ≥ 300 mg/kg/day** (HDT).

This study is classified as **Core-Guideline (Acceptable)** and satisfies the requirement, Guideline 83-3, for a developmental toxicity (teratology) study in rats.

Executive Summary for Developmental Toxicity Study in Rabbits
[Guideline No. 83-3b (870.3700), MRID 44610701]

In this rabbit developmental toxicity study (MRID 44610701) submitted as confirmatory data following the 1996 RED, Mepiquat chloride (56.7% a.i.) in doubly distilled water was administered to 15 pregnant Himalayan rabbits/dose by gavage on gestation days 7-19 in doses of 50, 100 or 150 mg/kg body weight/day.

In the mid-dose group, a reduction in body weight gains and food consumption observed only

during gestation days 11-14, was considered to be within biological variation and not of toxicological significance. In the high-dose, maternal toxicity was characterized by clinical signs of toxicity, reduced body weight gains, and reduced food consumption. Blood was observed in the bedding during post-treatment in 2/15 high-dose females. High-dose body weight gains were reduced from gestation days 11-16 ($p \leq 0.05$ or 0.01) and rebounded during the post-treatment interval (↑42%, not statistically significant, days 23-25). Additionally, decreases ($p \leq 0.05$ or 0.01) in food consumption of the high-dose dams were noted during treatment (↓31-68%, gestation days 8-20).

There were no treatment-related effects noted in mortality, gross pathologic findings, or cesarean section parameters at any dose level.

Maternal LOAEL = 150 mg/kg body weight/day based on reduced body weight gains and reduced food consumption.
Maternal NOAEL = 100 mg/kg body weight/day

There were no treatment-related external, visceral, or skeletal malformations or external and visceral variations noted at any dose level. However, increased incidences of skeletal variations such as epactal bone between nasal and frontal bones, accessory 13th rib(s), and accessory thoracic vertebra were observed. The following high-dose percent litter incidences were outside of historical control ranges: epactal bone between nasal and frontal bones (high-dose, 43%; historical control range, 0-30.0%); accessory 13th rib(s) (high-dose, 50%; historical control range 0-26.7%); and accessory throacic vertebra (high-dose, 14%; historical control range 0-13.3%).

Developmental LOAEL = 150 mg/kg/day based on increased skeletal variations.
Developmental NOAEL = 100 mg/kg/day

This developmental toxicity study is classified acceptable and fulfills the Guideline (83-3b). An explanation should have been submitted regarding the use of test material with an active ingredient content of only 56.7%.

4. <u>Reproductive Toxicity Study Conclusions</u>

<u>Executive Summary for 2-Generation Reproduction Study in Rats</u>
[Guideline No. 83-4 (870.3800), MRID 43378601]

Groups of 25 male and 25 female Wistar rats were fed mepiquat chloride in their diets at concentrations of 0, 500, 1500 or 5000 ppm for 10 weeks (F0) or 14 weeks (F1) before mating, and during mating, gestation, and lactation. The F0 parents were mated a second time 2 weeks after weaning the first litter. The doses corresponding to the dietary concentrations are 51.2 and 48.6, 153.1 and 146.6 and 499.3 and 574.5 mg/kg/day, respectively for F0 and F1 males and 54.0 and 53.3, 163.6 and 162.0, and 530.0 and 626.5 mg/kg/day, respectively for F0 and F1 females.

No treatment-related systemic effects occurred in male or female rats receiving 500 or 1500 ppm of the test material. In animals receiving 5000 ppm (high-dose), effects indicative of impaired

neurological function included tremors and hypersensitivity upon handling in 70-85% of F0 anf F1 dams; decreased forelimb strength (7 to 17%, $p<0.01$) and hindlimb grip strength (11 to 16%, $p<0.01$) in F0 and F1 dams before mating, during lactation or after weaning; and decreased hindlimb grip strength (15%, $p<0.01$) in high-dose F1 males. Mean body weights of the high-dose F0 males were reduced by 10-11% ($p<0.01$) relative to controls during the entire 29-week treatment period. Mean body weights of high-dose F1 males were reduced by about 50% relative to controls at the start of the premating period; thereafter, the animals steadily gained weight such that at the end of the premating period, overall body weight gain was only slightly reduced (15%). Food consumption in high-dose F1 males was reduced by 15% ($p<0.01$) compared with controls. Mean body weight of high-dose F0 females was reduced by 7% ($p<0.05$) and body weight gain by 11% ($p<0.05$) during the premating period. Gestation body weight of F0 females was reduced by 10% or less and body weight gain by 12% or less. High-dose F0 dams lost weight during lactation, and weight gain of their F1 pups was reduced by as much as 39% during the same period. High-dose F1 dams lost weight during lactation, and weight gain of the F2 pups was reduced by 36%. Food consumption by the high-dose dams was reduced by 31 to 33% during lactation. Changes in hematologic, clinical chemistry, and urinalysis parameters in the adult high-dose rats were unreleated to dose, biologically insignificant, or were due to the reduced body weight. Plasma, erythrocyte and brain cholinesterase activities were not affected by treatment with the test material. Decreased liver and kidney weights and decreased incidence of lipid storage in the liver observed in high-dose males and females were consistent with the decreased terminal body weights and are unlikely to be due to toxicity of the test material. A significant number of high-dose F1 and F2 pups were slow in reaching developmental milestones (pinna unfolding, opening of the auditory canal and eyes, and gripping reflex). These effects are attributed to retarded growth of the pups.

The LOAEL for parental systemic toxicity is 5000 ppm (HDT) for male and female rats based on neurological impairment, decreased body weight and body weight gain in the adults, and retarded growth of F1 and F2 pups. The corresponding NOAEL is 1500 ppm.

There were no treatment-related effects on reproductive parameters.

The LOAEL for offspring toxicity is 5000 ppm based on the significant number of high-dose F1 and F2 pups which were slow in reaching developmental milestones (pinna unfolding, opening of the auditory canal and eyes, and gripping reflex). The corresponding offspring NOAEL is 1500 ppm.

This study is classified as core-guideline (acceptable) and satisfies the requirements for a multigeneration reproduction feeding study (83-4).

5. Additional Information from Literature Sources

None

6. Pre-and/or Postnatal Toxicity

The HIARC concluded that there is not a concern for pre- and/or postnatal toxicity resulting from exposure to mepiquat chloride.

A. Determination of Susceptibility

Based on the results in a developmental toxicity study in rats and in a developmental toxicity study in rabbits, there was no quantitative or qualitative evidence of increased susceptibility of rat or rabbit fetuses to *in utero* exposure to mepiquat chloride.

Based on the results in a 2-generation reproduction study in rats, there was no quantitative or qualitative evidence of increased susceptibility of neonates (as compared to adults) to mepiquat chloride.

B. Degree of Concern Analysis and Residual Uncertainties

There are no concerns or residual uncertainties for pre and/or post-natal toxicity following exposure to mepiquat chloride.

C. Special FQPA Safety Factor(s):

Based upon the above-described data, no special FQPA safety factor is needed (i.e. 1X) since there are no residual uncertainties for pre and/or post natal toxicity.

The Special FQPA Safety Factor recommended by the HIARC **assumes** that the exposure databases (dietary food, drinking water, and residential) are complete and that the risk assessment for each potential exposure scenario includes all metabolites and/or degradates of concern and does not underestimate the potential risk for infants and children.

7. Recommendation for a Developmental Neurotoxicity Study

The HIARC concluded that there is a concern for developmental neurotoxicity resulting from exposure to mepiquat chloride.

On August 31, 1999, HIARC recommended that a developmental neurotoxicity study in rats be conducted with mepiquat chloride based on the following concerns:

- Mepiquat chloride is a known neurotoxicant with a known mechanism of action (nicotinic ganglionic stimulant). It shows a structure-activity-relationship for neurotoxicity and there is concern for a possible increase in severity of neurotoxic effects in developing offspring.
- In the reproduction toxicity study, a significant number of high-dose pups were slow in reaching developmental milestones: pinna unfolding; opening of the auditory canal and eyes; and gripping reflex. It was stated that the effects may be more severe in the pups due to the developing blood-brain barrier.

On January 22, 2003, based on the weight of evidence presented, the HIARC reaffirmed the previous conclusion that a developmental neurotoxicity (DNT) study conducted with mepiquat chloride in rats is required. HIARC determined that a 3X (as opposed to 10X) database uncertainty factor (UF_{DB}) is adequate to account for the lack of the DNT study based on the following considerations:

- It is assumed that the DNT study will be conducted at dose levels similar to those used in the rat reproduction study (0, 48.6, 146.6, and 499.3 mg/kg/day) wherein the offspring NOAEL / LOAEL was 146.6 / 499.3 mg/kg/day, respectively.
- All of the toxicity endpoints for risk assessment (acute and chronic Reference doses, short- and intermediate-term dermal and inhalation endponts) are based on the NOAEL of 58.4 mg/kg/day established from the combined results of the 1-year dog feeding study and the 90-day dog feeding study (LOAEL is 170 mg/kg/day based on salivation).
- A comparison of the results from the rat reproduction study with the dog studies indicates that the dog is the more sensitive species by a factor of approximately 3X.
- The results of the DNT study would not be expected to impact the current regulatory dose since the NOAEL used for all risk assessment scenarios for mepiquat chloride (58.4 mg/kg/day) is 3-fold lower than the parental or offspring NOAEL observed in the rat reproduction study (approximately 150 mg/kg/day).
- Although the DNT study will be conducted in the rat (and not the dog), the apparent sensitivity of the dog is accounted for in the 10X Interspecies factor.

Although the results of the DNT are not expected to impact the current regulatory dose given the 3-fold difference observed in the rat and dog studies, HIARC does not have sufficient reliable data to apply no additional safety factor. Rather, HIARC believes that the 3X difference between the rat and dog studies, provides reliable data supporting a 3X reduction in the default 10x factor. The use of a 3X will provide roughly a 10X difference between the NOAEL associated with the identified effects in the rat necessitating the DNT study and the NOAEL from the dog study used for setting regulatory doses. Therefore, a UF_{DB} of 3X will be applied to account for the lack of the DNT study with mepiquat chloride. It is also noted that the 3X database uncertainty factor is also adequate to account for the lack of the required acute and subchronic neurotoxicity studies with mepiquat chloride since these studies will be conducted in adult animals at doses higher than (acute) or comparable to (subchronic) those tested in the developmental neurotoxicity study.

II. HAZARD IDENTIFICATION

1. Acute Reference Dose (RfD) - General Population

Study Selected: One-year dog feeding study; 90-day dog feeding study § 870.4100; 870.3150

MRID Nos.: 41488105 and 43264403 (One year); 00135720 (90-day)

Executive Summary: In a chronic toxicity study (MRID 414488105), Reg. No. 85 559, Mepiquat chloride (99.5% a.i.) was administered to 6 beagle dogs/sex/dose in the diet at dose levels of 0, 200, 600 or 1800 ppm (0, 6.3, 19.9 or 58.4 mg/kg/day, respectively) for 12 months.

The only possible treatment-related effect, observed at the 1800 ppm dose, was a very slightly increased storage of the iron pigment in the spleen of 3 male dogs and in the liver of 2 male dogs. All of the remaining male and female dogs in this group, and all of the males and the majority of the females in the remaining groups, including the controls, also had iron pigment in the spleen and liver, but of slightly lesser severity (Grade 1). There were no compound-related effects on mortality, clinical signs, body weight, food consumption, ophthalmoscopic findings, hematology, clinical chemistry, urinalysis, organ weights, gross pathology and, with the exception of the iron pigment noted above, histologic pathology.

Since no definite toxic effects were detected in the current study (MRID 41488105), another one-year dog feeding Supplementary Study (MRID 43264403) with two doses of mepipuat chloride (0 and 6000 ppm) was started in January, 1992 and completed on May 5, 1994. Both studies were, therefore, considered in establishing a NOAEL and determining whether the guideline requirement 83-1b was satisfied.

Based on both studies (MRIDs 41488105 and 43264403), the LOAEL for males and females is 6000 ppm (170 mg/kg/day) and the NOAEL is 1800 ppm (58.4 mg/kg/day). Toxic signs observed at the 6000 ppm dose in all dogs were: (1) impaired neurological functions (salivation after feeding and, in one female, lateral position, extension spasm and ataxia of the hind limb); (2) epithelial vacuolization of the renal distal tubules; and (3) increased hemosiderin in the spleen (males only). Considered together with another study (MRID 43264403), this chronic toxicity study in the dog (MRID 41488105) is acceptable and satisfies the guideline requirement for a chronic oral study (83-1b) in dogs.

In a chronic toxicity supplementary study (MRID 43264403), Mepiquat chloride, 56.05% a.i. (w/w) in water, was administered to 6 beagle dogs/sex/dose in diet at dose levels of 0 and 6000 ppm (170 mg/kg/day) for 12 months. The following effects were observed in the treated group: salivation in all dogs; early mortality (one female was sacrificed moribund on study day 17); kidney vacuolization in 4/6 males (controls: 1/6) and 5/6 females (controls: 2/6); and increased hemosiderin storage in spleen of male dogs. Salivation (an indicator of impaired neurological functions) occurred at 2 hours after each feeding, was slight at first, moderate to severe during the next 4 hours and then gradually disappeared. The moribund dog showed weakness and ataxia of the hind legs, lateral position, extension spasm, abnormal body temperature (no details), stomach lesion, lung focus and cyst in the pituitary gland. The kidney vacuolization was minimal to slight in the treated dogs and minimal in the controls. The hemosiderin storage in the spleen of the male dogs showed a higher intensity in the treated group than in the controls.

This study should be considered together with an earlier study (MRID 41488105) in which 3 doses of Mepiquat chloride were tested (200, 600 or 1800 ppm, equivalent to 6.3, 19.9 or 58.4

mg/kg/day, respectively), but in which a definitive LOAEL was not determined. Based on both studies, the LOAEL for males and females is 6000 ppm (170 mg/kg/day) and the NOAEL is 1800 ppm (58.4 mg/kg/day). The guideline requirement for a chronic feeding study (83-1b) in dogs is satisfied. By itself, the current study is classified as Supplementary and does not satisfy the 83-1b data requirement.

In a subchronic toxicity study (MRID 00135720), technical Mepiquat chloride (no purity given) was administered to 4 beagle dogs/sex/dose in the diet at dose levels of 0, 100, 300, 1000 or 3000 ppm (0, 3.3, 9.8, 32.4 or 95.3 mg/kg/day). The LOAEL is 3000 ppm (95.3 mg/kg/day), based on clinical signs of toxicity (slight sedation: tonic/clonic spasms were noted while animals were lying on their abdomen or side); inhibition of body weights (up to 14% less); and hematological effects (up to 14% reduction in hemoglobin content, number of erythrocytes and reduced hematocrit). The NOAEL is 1000 ppm (32.4 mg/kg/day). This subchronic toxicity study is classified as supplementary (upgradable pending receipt of additional information on test material, particularly purity). It does not satisfy the guideline requirement for a subchronic oral study (82-1) in the dog.

Dose and Endpoint for Establishing RfD: NOAEL = 58.4 mg/kg based on salivation seen 2 hours post treatment.

Uncertainty Factor (UF): 300 (10X for interspecies extrapolation, 10X for intraspecies variations,and 3X for an incomplete database for lack of developmental neurotoxicity study)

Comments about Study/Endpoint/Uncertainty Factor: The endpoint of concern (salivation) is an indicator of impaired neurological function and was observed at 2 hours after each feeding. Salivation was slight at first, moderate to severe during the next 4 hours and then gradually disappeared. Since the endpoint of concern was seen during the first 4 hours of dosing, this was deemed appropriate for this (acute) dietary risk assessment. This endpoint was also supported by sedation (also a neurotoxic sign) which was observed 1 to 6 hours post dosing in the subchronic study.

Due to its quarternary nitrogen, Mepiquat chloride is not expected to readily pass the blood-brain barrier and is virtually devoid of CNS effects. Most of the acute effects described in animal studies are due to its action on the peripheral nervous system. It's prominent action is at the ganglionic receptor sites, similar to nicotine. Like nicotine, the onset of symptoms causes an initial stimulation of salivation and bronchial secretions followed by muscle weakness or paralysis. In the dog studies, salivation following each feeding is most likely due to the accumulation of unswallowed saliva in the pharynx.

Acute RfD (General Population) = $\frac{\text{NOAEL: 58.4 mg/kg}}{\text{UF: 300}}$ = **0.195 mg/kg**

2. Chronic Reference Dose (cRfD)

Study Selected: One-year dog feeding study; 90-day dog feeding study § 870.4100; 870.3150

MRID Nos.: 41488105 and 43264403 (One year); 00135720 (90-day)

Executive Summary: See Acute RfD

Dose and Endpoint for Establishing RfD: NOAEL of 58.4 mg/kg/day based on clinical signs of toxicity (sedation, abdominal and lateral positioning and tonic/clonic spasms), decreased body weight and hematological changes were observed at the LOAEL of 95.3 mg/kg/day.

Uncertainty Factor(s): 300 (10X for interspecies extrapolation, 10X for intraspecies variations,and 3X for an incomplete database for lack of developmental neurotoxicity study)

Comments about Study/Endpoint/Uncertainty Factor: The HIARC concurred with the study/dose/endpoints selected by the RfD Committee (5/2/96) which recommended that the RfD for this chemical be based on the combined chronic and subchronic toxicity studies in dogs with a NOAEL of 58.4 mg/kg/day and a LOAEL of 95.3 mg/kg/day.

Chronic RfD =	NOAEL: 58.4 mg/kg/day UF: 300	=	**0.195 mg/kg/day**

3. Incidental Oral Exposure: Short-Term (1-30 days) and Intermediate-Term (1 - 6 Months)

These endpoints were not addressed in the original HIARC evaluation in 1999 (see TXR NO. 013809).

4. Dermal Absorption

Dermal Absorption Factor: 25%

There were no dermal absorption data available. Therefore, comparisons between an oral study and a dermal study could not be made. A dermal absorption factor was extrapolated using the acute studies.

Acute Oral Toxicity in Rats (MRID 41488101): The doses tested were 100, 200, 464, 1470 and 2150 mg/kg. The LD50 was 464 mg/kg (males and females). Toxicity reported at ≥464 mg/kg included poor general health, dyspnea, apathy, staggering, twitching, compulsary gnawing and cyanosis. There was general congestion in the nonsurvivors.

Acute Dermal Toxicity in Rats (MRID 41488102): The single dermal dose was 2000 mg/kg. The

LD50 was >2000 mg/kg. There were no clinical signs observed.

The amount absorbed was determined to be 25% based on the comparison of these two studies. Therefore, the dermal absorption factor is extrapolated to be 25% (464 ÷ 2000 x 100 = ≈25%). This would provide a conservative dermal absorption value since no dermal toxicity was seen in a dermal LD50 study.

5. Dermal Exposure: Short-Term (1 - 30 days) and Intermediate-Term (1 - 6 Months)

Study Selected: One-year dog feeding study; 90-day dog feeding study § 870.4100; 870.3150

MRID Nos.: 41488105 and 43264403 (One year); 00135720 (90-day)

Executive Summary: See Acute RfD

Dose and Endpoint for Establishing RfD: NOAEL of 58.4 mg/kg/day based on clinical signs of toxicity (sedation, abdominal and lateral positioning and tonic/clonic spasms), decreased body weight and hematological changes were observed at the LOAEL of 95.3 mg/kg/day.

Comments about Study/Endpoint: An oral dose was selected due to the lack of a dermal toxicity study. A dermal absorption factor of 25% should be used for risk assessment. This endpoint is appropriate for short- and intermediate-term since the NOAEL in the one-year study was 1800 ppm (58.4 mg/kg/day) and was based on salivation in all dogs, weakness and ataxia in the hind legs, lateral position, stomach lesion and lung foci in one female sacrificed moribund on study day 17. The following toxic signs were observed in the 90-day feeding study at the 3000 ppm level (95.3 mg/kg/day; LOAEL), after 4 weeks (or longer) of treatment: sedation, tonoclonic spasms, abdominal and lateral positions and paralysis.

6. Dermal Exposure: Long-Term (> 6 Months)

This endpoint was not addressed in the original HIARC evaluation in 1999 - it was determined that considering the use pattern (foliar applied; one application/year) of this plant growth regulator, a chronic occupational or residential exposure by the dermal route is unlikely (see TXR NO. 013809).

7. Inhalation Exposure: Short-Term (1 - 30 days) and Intermediate-Term (1 - 6 Months)

Study Selected: One-year dog feeding study; 90-day dog feeding study § 870.4100; 870.3150

MRID Nos.: 41488105 and 43264403 (One year); 00135720 (90-day)

Executive Summary: See Acute RfD

Dose and Endpoint for Establishing RfD: NOAEL of 58.4 mg/kg/day based on clinical signs of toxicity (sedation, abdominal and lateral positioning and tonic/clonic spasms), decreased body weight and hematological changes were observed at the LOAEL of 95.3 mg/kg/day.

Comments about Study/Endpoint: Except for an acute inhalation toxicity study, no inhalation studies are available. Therefore, the HIARC selected oral NOAELs for inhalation risk assessments. An inhalation absorption factor of 100% (default value assuming equivalent inhalation and oral absorption) will be used for route-to-route extrapolation.

8. Inhalation Exposure: Long-Term (> 6 Months)

This endpoint was not addressed in the original HIARC evaluation in 1999 - it was determined that no long term exposure would occur with current proposed registrations (see TXR NO. 013809).

9. Margins of Exposure

Summary of target Margins of Exposure (MOEs) for risk assessment.

Route / Duration	Short-Term (1-30 Days)	Intermediate-Term (1 - 6 Months)	Long-Term (> 6 Months)
Occupational (Worker) Exposure			
Dermal	100	100	NA
Inhalation	100	100	NA
Residential (Non-Dietary) Exposure			
Oral	NA	NA	NA
Dermal	NA	NA	NA
Inhalation	NA	NA	NA

For Occupational exposure: short-term (1 day to 1 month) and intermediate-term (1 to 6 months) dermal and inhalation risk assessments, a MOE of 100 is required. This is based on the conventional uncertainty factor of 100X (10X for intraspecies extrapolation and 10X for interspecies variation). **Long-term (>6 months) exposure is not expected.**

For Residential exposure: No residential use. MOEs are not applicable (NA)

10. Recommendation for Aggregate Exposure Risk Assessments

There are no residential uses. Therefore, aggregate exposure risk assessment is not required.

III. CLASSIFICATION OF CARCINOGENIC POTENTIAL

1. Combined Chronic Toxicity/Carcinogenicity Study in Rats

MRID No.: 43264402

Executive Summary: In a chronic feeding study, Mepiquat chloride was administered for 24 months in the diet to 20 Wistar rats/sex/dose at concentrations of 0, 290, 2316 and 5790 ppm (active ingredient), equivalent to doses of 0, 13, 106, 268 mg/kg/day for males and 0, 18, 146 and 371 mg/kg/day for females, respectively. Total food consumption for rats in the high (5790 ppm) and medium (2316 ppm) dose groups was decreased 8% and 1% (not significant), respectively, for males and 6% and 2%, respectively, for females relative to controls. Body weights for rats in the high dose group were decreased 12% for males and 12% for females (not significant), relative to controls at day 728 of the treatment period. Body weight gains from day 0 to 728 were decreased relative to controls for males (17%) and females (20%, not significant) in the high dose group. There was an increase in the macroscopic pathological finding of focus for the adrenal cortex (control: 9/20; 5790 ppm: 16/20) and in microscopic pathological findings of vacuolated cell foci (control: 3/20; 5790 ppm: 9/20), hemosiderin pigment, and nodular hyperplasia for the adrenal cortex for females in the high dose group. There was an increased incidence of urinary crystals for males in the high dose group (12/20) as compared to controls (4/20) at day 99 and the increased incidence persisted (not significant) through day 541 of the treatment period. The NOAEL is 2316 (active ingredient). The LOAEL is 5790 ppm (active ingredient) based upon decreased body weights and body weight gains for males and females, increases in urinary crystals for males and pathological changes in the adrenal cortex in females. This study satisfies the guideline requirements for a chronic feeding study in rats (83-1a).

MRID No.: 43396001

Executive Summary: In a carcinogenicity study, Mepiquat Chloride was administered for 24 months in the diet to 50 Wistar rats/sex/dose at concentrations of 0, 290, 2316 and 5790 ppm (active ingredient), equivalent to doses of 0, 13, 105 and 269 mg/kg/day for males and 0, 17, 141 and 370 mg/kg/day for females, respectively. For male and female rats treated with Mepiquat Chloride at 5790 ppm, group mean daily and total food consumption were 88% ($p<0.01$) and 93% ($p<0.01$) of controls, respectively, group mean body weights at day 728 were 82% ($p<0.01$) and 81% ($p<0.01$) of controls, respectively, group mean body weight gains from day 0 to day 728 were 77% ($p<0.01$) and 71% ($p<0.01$) of controls, respectively and food efficiency was 90% ($p<0.05$) and 78% ($p<0.01$) of controls, respectively. For males and females in the 5790 ppm dose group relative to controls, there were macroscopic pathological findings for the adrenal cortex (focus: 5/50 vs. 16/50, $p<0.01$ for males; 31/50 vs. 39/50, $p<0.05$ for females) and the ovaries (cyst: 14/50 vs. 24/50, $p<0.05$), and non-neoplastic pathological findings for the liver

(biliary cysts: 8/50 vs. 16/50, $p<0.05$ for females), the kidneys (tubular casts: 6/50 vs 13/50, $p<0.05$ for females; tubular atrophy: 5/50 vs. 12/50, $p<0.05$ for males), the prostate (alveolar atrophy: 1/50 vs. 7/50, $p<0.05$), the ovaries (dilated bursa: 1/50 vs. 13/50, $p<0.01$), the uterus (stromal hyperplasia: 2/50 vs. 11/50, $p<0.01$; squamous hyperplasia: 5/50 vs. 14/50, $p<0.05$), the sublingual glands (acinar atrophy: 4/49 vs. 14/50, $p<0.01$), and the mammary glands (secretion: 30/47 vs. 45/50, $p<0.01$). There were no toxicologically significant findings for the medium- or low-dose groups. There was no excess mortality for treated rats relative to controls. The LOAEL for males and females is 5790 ppm, based upon decreased body weights, body weight gains, food consumption, food efficiency, and macroscopic and non-neoplastic findings. The NOAEL for males and females is 2316 ppm. There were no treatment-related neoplastic findings for males or females treated with Mepiquat chloride. Thus, Mepiquat Chloride does not exhibit carcinogenic potential in a 2-year feeding study involving male and female Wistar rats over this dose range. Based upon the decreased body weights and body weight gains, 5790 ppm is a maximum tolerated dose (MTD) for Mepiquat Chloride for 2-year feeding to male and female Wistar rats, but is adequate for identifying carcinogenic potential. This study satisfies the guideline requirements for an ocnogenicity study in rats (83-2a).

2.Carcinogenicity Study in Mice

MRID No. 43264404

Executive Summary: In a carcinogenicity study, Mepiquat Chloride was administered in the diet for 24 months to 50 B6C3F1/CrlBr mice/sex/dose and for 12 months to 10 mice/sex/dose at concentrations of 0, 500, 2000 and 7500 ppm (active ingredient). For main group mice, these respective doses are equivalent to 0, 74, 297 and 1140 mg/kg/day for males and 0, 85, 328 and 1348 mg/kg/day for females, averaged over the 24-month feeding study. There were no treatment-related effects of Mepiquat Chloride administration on group mean body weights or body weight gains over the 24 month treatment period. There were no treatment-related macroscopic or non-neoplastic microscopic pathological findings for males or females. The NOAEL for Mepiquat Chloride administered for 2 years in food is 7500 ppm for male and female B6C3F1 mice. There were no treatment-related neoplastic findings for males or females treated with Mepiquat Chloride. Thus, Mepiquat Chloride does not exhibit carcinogenic potential in a 2-year feeding study involving male and female B6C3F1 mice over this dose range. Based upon the lack of treatment-related findings, Mepiquat Chloride was not administered at the MTD. However, the high dose (7500 ppm) for the study was sufficient to assess carcinogenicity since the limit dose of 1000 mg/kg/day was exceeded. This study satisfies the guideline requirements for an oncogenicity study in mice (83-2a).

There was no evidence of carcinogenicity in male or female mice. The incidence of neoplasms found in numerous organs and tissues was comparable in treated and control groups. The animals were dosed adequately since their intake exceeded the limit dose of 1000 mg/kg/day.

Classification of Carcinogenic Potential: The HIARC agreed with the conclusions of the RfD Committee that Mepiquat chloride should be classified as a "**not likely**" human carcinogen

according to the EPA *Proposed Guidelines for Carcinogen Risk Assessment* (April 10, 1996). This classification is based on the lack of evidence of carcinogenicity in male and female rats, in male and female mice and on the lack of genotoxicity in an acceptable battery of mutagenicity studies.

IV MUTAGENICITY

There were four acceptable mutagenicity studies:

(a) Salmonella typhimurium reverse gene mutation assay (MRID 41488106): The test is negative in all strains in the presence or absence of metabolic activation system (+/-S9) up to 2500 µg/plate, the highest dose tested.

(b) Chinese hamster ovary (CHO) cell chromosome aberration assay (MRID 41488107): The test is negative up to 5000 µg/mL, the highest dose tested, with or without metabolic activation.

(c) Mouse dominant lethal assay (MRID 00071948: The test is negative in male mice up to 3000 ppm, the highest dose tested, administered once daily for 5 consecutive days by oral gavage (≈450 mg/kg/day). There was, however, no overt toxicity or adverse effects on reproductive or dominant lethal parameters.

(d) In vitro unscheduled DNA systhesis in primary rat hepatocytes (MRID 41488108): The test is negative up to cytotoxic concentrations (≥4000 µg/mL).

The RfD Committee (May 2, 1996), "concluded that neither the bacterial gene mutation assay nor the mouse dominant lethal assay evaluated a maximum tolerated dose or a limit dose. It was concluded, however, that Mepiquat chloride was adequately tested and that it is not likely that testing higher dose levels or concentrations would have altered the negative response. This assessment was based on the negative findings of the other mutagenicity studies submitted and in conjunction with the lack of an oncogenic effect in rat or mouse long-term feeding studies. Additionally, the absence of significant reproductive or developmental toxicity attributable to a mutagenic mode of action (i.e., decreased total implants, increased resorptions) supports this conclusion. The acceptable studies satisfy the pre-1991 mutagenicity initial testing battery guidelines. Based on the available toxicology data, there is no concern for mutagenicity at this time."

V. HAZARD CHARACTERIZATION

The toxicology studies conducted on mepiquat chloride demonstrate that, other than decreases in body weight and/or body weight gains and/or food consumption, the primary effect that the chemical has is as

a neurotoxicant. Observations regarding neurotoxicological parameters were noted in multiple species, both sexes and in adult animals in addition to rat pups.

VI. DATA GAPS / REQUIREMENTS

Acute neurotoxicity study - rats
Subchronic neurotoxicity study - rats
Developmental neurotoxicity study - rats

VII. ACUTE TOXICITY

Acute Toxicity of Mepiquat chloride

Guideline No.	Study Type	MRID No.	Results	Toxicity Category
81-1	Acute Oral, Rat	41488101	LD50 = 464 mg/kg	II
81-2	Acute Dermal, Rat	41488102	LD50 = > 2000 mg/kg	III
81-3	Acute Inhalation, Rat	41954101	LC50 = > 4.89 mg/L	IV
81-4	Primary Eye Irritation, Rabbit	00071942	Not irritant; Score: 1.4/110	IV
81-5	Primary Skin Irritation, Rabbit	41488103	Not irritant; Score: 0/4	IV
81-6	Dermal Sensitization, G. Pig	41488104	Negative	-
81-8	Acute Neurotoxicity, Rat	(a)	-	-

(a) = Because of the limited use of Mepiquat chloride, low application rates and the findings that Mepiquat chloride was neurotoxic in several studies with rats (81-1, 82-1a, 83-3a and 83-4) at high levels only (300-889 mg/kg/day), the acute neurotoxicity study was not required. However, the acute neurotoxicity study in the rat (as well as the subchronic neurotoxicity study in the rat and the developmental neurotoxicity study in the rat) is in the process of being proposed in a Data Call In.

VIII. SUMMARY OF TOXICOLOGY ENDPOINT SELECTION

Summary of Toxicology Endpoint Selection for Mepiquat chloride

Exposure Scenario	Dose Used in Risk Assessment, UF	Special FQPA SF* and Level of Concern for Risk Assessment	Study and Toxicological Effects
Acute Dietary (General Population including infants and children)	NOAEL = **58.4 mg ai/kg/day** UF =**300** **Acute RfD** = **0.195** mg/kg/day	FQPA SF = **1X** **aPAD** = acute RfD / FQPA SF = **0.195** mg/kg/day	1-year dog feeding; 90-day dog feeding Salivation (indicator of impaired neurological function) at the LOAEL of 95.3 mg/kg/day.
Chronic Dietary (All populations)	NOAEL = **58.4 mg ai/kg/day** UF =**300** **Chronic RfD** = **0.195** mg/kg/day	FQPA SF = **1X** **cPAD** = chronic RfD / FQPA SF = **0.195** mg/kg/day	1-year dog feeding; 90-day dog feeding Salivation (indicator of impaired neurological function) at the LOAEL of 95.3 mg/kg/day.
Short-Term (1 - 30 days) and **Intermediate-Term Dermal** (1 - 6 months)	Oral NOAEL = **58.4 mg ai/kg/day**[a]	**Residential** MOE = **NA** **Occupational** MOE = **100**	1-year dog feeding; 90-day dog feeding Salivation (indicator of impaired neurological function) at the LOAEL of 95.3 mg/kg/day.
Long-Term Dermal (>6 months)	None.	**Residential** MOE = **NA** **Occupational** MOE = **NA**	Not selected.
Short-Term (1 - 30 days) and **Intermediate-Term** (1 - 6 months) **Inhalation**	Oral NOAEL = **58.4 mg ai/kg/day**[b]	**Residential** MOE = **NA** **Occupational** MOE = **100**	1-year dog feeding; 90-day dog feeding Salivation (indicator of impaired neurological function) at the LOAEL of 95.3 mg/kg/day.
Long-Term Inhalation (>6 months)	None	**Residential** MOE = **NA** **Occupational** MOE = **NA**	Not selected.

a = 25% dermal absorption factor should be used in risk assessment.
b = Since oral values were selected 100% inhalation absorption factor (default) value should be used in route-to-route extrapolation/risk assessment.

UF = uncertainty factor, FQPA SF = Special 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, NA = Not Applicable

NOTE: The Special FQPA Safety Factor recommended by the HIARC **assumes** that the exposure databases (dietary food, drinking water, and residential) are complete and that the risk assessment for each potential exposure scenario includes all metabolites and/or degradates of concern and does not underestimate the potential risk for infants and children.