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

MAR 18, '81

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OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

Caswell #215B

DATE:

SUBJECT: Proposal to establish additional tolerances for Chlorothalonil (Daconil; DAC-2787; DCN) and its 4-OH metabolite (DAC-3701):

PP# 6F1799 - 0.2 ppm in soybeans
PP# 1G2428/1H5278 - 0.1 ppm in citrus; 10 ppm in citrus oil
PP# 0F2405/0H5272 - 0.1 ppm in oranges and grapefruit; 10 ppm in Citrus Oil

PP# 6E1887 - 1.0 ppm on parsnips (minor crop tolerance)

FROM: David Ritter, Toxicologist DR 3/4/81
Review Section #1
Toxicology Branch/HED (TS-769)

TO: Henry Jacoby, PM #21
HFB/RD (TS-767)

Clinton Fletcher, Minor Use Officer
PCB/RD (TS-767)

Petitioner: Diamond Shamrock
Cleveland Ohio
Soybeans and Citrus

IR-4 Regional Coordination
New Brunswick, N.J.
Parsnips

1-23

Conclusions

1. Soybeans - The proposed tolerance of 0.2 ppm combined residues of chlorothalonil (Daconil) is safe and will protect the public health.

2. Citrus and Citrus Oil - 0.1 and 10 ppm respectively. The proposed tolerances are safe and will protect the public health.

   NOTE: This comment applies to the proposal only, for the purpose of estimating relative exposure and risk from these tolerances. The RCB (review of 1/30/81, L. Bradley) has concluded that the proposed tolerance levels may not be adequate due to problems with the analytical methodology and lack of sufficient residue data in oranges and grapefruit to estimate an appropriate tolerance.

3. Parsnips - The proposed minor crop tolerance in parsnips is safe and will protect the public health.

   NOTE: We defer to RCB the question of meat, eggs and milk residues that may result from animal feed use of soybeans and its animal feed byproducts and from citrus animal feed byproducts.

Bases for the Conclusions

1. Incremental risk and exposure analyses show that the proposed tolerances would not exceed the Upper Limit on Risk (ULR) at 10^{-6} or 1/1,000,000, based on a Cancer study in rats reported by NCI. Dietary residues theoretically would not be significantly increased [i.e., less than 1% of the existing Theoretical Maximum Residue Contribution (TMRC)] over theoretical residues postulated from previously established tolerances.

2. The Maximum Permissible Ingestion (MPI) has been established at 0.90 mg/day/60 kg based on chronic feeding studies in the dog with a systemic NOEL of 60 ppm. This will not be exceeded by the addition of these tolerance. Tolerances for residues of CTN and its 4-OH metabolite* have been established pursuant to 40 CFR 180.275 for a number of human food items. These are supported by numerous chronic feeding studies; reproduction studies and subacute and acute exposure studies using both the technical grade product of commerce and registered formulations. (See the free-standing summary sheet, attached, and the "One-liner" summary sheet for new data, attached).

*Our records indicate that the 4-OH metabolite is not a major metabolite in laboratory animals; however, it is a substantial portion of the total residues in plants. (Review of B. Malone, PP7F0599, 8/28/67).

3. HCB residues in soybeans are expected to be less than 0.003 - 0.006 ppm. HCB residues in parsnips are expected to be less than 0.04 ppm*. These levels are not of toxicological concern. We have no estimates of HCB residues in citrus or citrus oil (See #2.).

Detailed Considerations

1. Petitioner has submitted an independent analysis of National Cancer Institute's (NCI) bioassay of CTN for carcinogenicity (NCI-CG-TR-41; 1978). NCI has concluded that CTN was carcinogenic in male and female Osborne-Mendel rats, but not in B6C3F1 mice. Toxicology Branch, on the basis of this assay, disapproved a minor crop tolerance in turnips of 1 ppm (D. Ritter review of 1/14/80, PP#7E1887). No attempt was made to validate the assay.

   The submitted analysis (by Booz-Allen and Hamilton, Inc.) compares data included in the published NCI report with raw data submitted to NCI by Tracor-Jitco, the prime contractor, and its sub-contracting laboratory, Gulf South Research Institute (GSRI). All raw data, lab books and correspondence relevant to the study were obtained from NCI under the Freedom of Information Act. Noteworthy findings include the following:

   - Only ten rats per sex were used in the control groups;
   - Much detailed analytical data was not available and is presumed to no longer exist:
   - There was a dispute between NCI and GSRI as to whether the lesions noted by the performing laboratory, GSRI, were in fact tumors and whether they were present in sufficient numbers to perform a statistical evaluation.

   Therefore since the integrity of the NCI study is in question, we are referring the matter to our Branch Pathologist for an in-depth review of both the NCI study and Booz-Allen's analysis of it.

*Below the detectable limit.

For the purpose of these petitions, however, we are assuming that the NCI study is accurate and that CTN is a rat carcinogen, albeit a very weak one. To this end we have prepared a carcinogenic Risk Analyisis as well as an Incremental Risk Assesment based on carcinogenic and chronic effects. These show that the theoretical exposure and risk associated with these proposed tolerances is less than one percent of the theoretical exposure from existing tolerances, and that the carcinogenic Upper Limits on Risk are less thatn 10^{-6} for current tolerances and is less that 10^{-7} for proposed tolerances. These considerations may be summarized as follows:

RAC	TMRC(1) mg/day/1.5 kg	% increased TMRC Increm./Priorx100	% of ADI	Upper Lim. Risk(3)
Prior Tols.	0.7007 "	-------	77.9%	$< 10^{-6}$
Soybeans	0.0028 "	0.39%	0.31%	$< 10^{-7}$
Citrus(2)	0.0057 "	0.54%	0.42%	$< 10^{-7}$
Citrus Oil(2)	0.0005 "	0.07	0.03%	$< 10^{-7}$
Parsnips	0.0004 "	0.06	0.033%	$< 10^{-7}$

(1) Theoretical Maximum Residues Contribution; See Table I for the complete analysis.

(2) Values for citrus are based on the submitted proposal. RCB has concluded that there are insufficient data as yet to characterize actual residues, Additional analytical methodology is needed (review of L. Bradley, 1/30/81).

(3) No attempt is made to extrapolate these values from rats to man.

From the preamble to the "Guideline for Conditional Registration" (FR 44:93; 5/11/79), we note that "...no additional tolerances will be granted when a significant data gap exists unless the incremental residue contribution is insignificant, generally less than one percent. This principle will apply to all tolerance actions (OGC memo of 6/13/79, C. Jablon, PP#7E1908/7E1914). These tolerances fall within this definition.

The Agency's policy with respect to oncogenic risk is embodied in the proposed "N-Nitrosamine" policy document of FR 45:124, 6/24/80, which sets a theoretical upper-limit-on-risk of one in a million (10^{-6}) as a reasonable regulatory criterion. The tolerances fall within this definition as well.

2. Additional comments as to the significance of histopathological lesions of the kidney in chronic and subacute feeding studies in the dog and rat were re-submitted. These comments had been submitted and were considered in support of previous petitions and include:

Hazelton # 200-175 (two year dog feeding study)

" 200-206 (two year dog feeding study)

" 200-205 (two year rat feeding study)

" 200-148 (two year rat feeding study)

" 200-175 (rat 76 week feeding study)

" 100-198 (4 month rat feeding study)

Bio-Tox 24-201 (4 month rat feeding study)

Petitioner has not indicated why this information is being resubmitted. Relevant studies involved the chronic feeding of CTN to the rat and dog. Dr. E. Long, Staff pathologist, determined that the definitive studies noted above were #200-206, dog, and 200-205, rat. She concluded that the no-effect level based on renal effects was 60 ppm in both studies. Each study showed definite compound-related renal pathology at the next higher (or highest) level of 120 ppm.

The 60 ppm NOEL in the dog was used to calculate the ADI of 0.015 mg/kg/day since the dog is the most sensitive species. (See the E. Long review of 1/26/76; 2/28/74 and 2/15/73, PP#2F1230).

3. A number of previously unsubmitted toxicity and mutagenicity studies accompany the petition and are herein examined:

a. Mutagenicity assays

i. Cell Transformation assay in newborn Fischer rats.

Identification: DTX-77-0037; 10/6/78

Laboratory: Microbiological Associates (Micro. Assoc.)

Substance Tested: DS-2787 (technical Daconil) 96% pure

Cell Lines: F1706 and H4536P+2

Summary:

10^{-3} ppm DS-2787 in the culture medium was the highest non-toxic dose that permitted sub-culturing of the treated cell lines for the transformed phenotype (ability to form macro-colonies in semisolid agar). Subcultures were then injected into new-born Fischer rats. Cells exposed to methylcholanthrene (MCA) served as positive controls.

At three months two rats from each litter were killed and examined for neoplasia.

Results:

No lesions were reported in rats whose cell lines were exposed to DS-2787. Tumors were reported for rats whose cells had been expose to MCA.

Conclusions:

Daconil does not induce phenotypic transformations in F1706 and H4536P+2 cell lines, nor do Daconil-treated cells induce neoplasia in newborn Fishcer mice upon injection thereof.

Note:

The relevance of *in vitro* mutagenicity testing has not been ascertained for regulatory purposes.

ii. DNA in Salmonella typhimurium (ST)

Identification: DTX-77-0033; 6/29/77

Laboratory: Micro. Assoc. Bethesda Maryland

Substance Tested: DAC 2787, 97.8% pure

Cell lines: ST; TA-1538 and TA-1978 (repair deficient and competent)

Summary:

DAC-2787 was tested at 2 - 20 ugm/plate with and without microsomal activiation (9-G rat liver supernatatant induced by Araclor). 6-Aminochrysens (6-AC) and 4-nitroquinoline-N-oxide (NQN) served as positive controls while Chloramphenicol (CAP) served as negative contro

Results:

A significant difference was noted between zones of inhibition for daconil and positive and negative controls in the TA-1538 strain. Differences for TA-1978 were suggestive but not definitive).

Conclusion:

Under the conditions of the test DAC-2728 interferes with the DNA repair mechanism of the TA-1538 (possitive results were suggestive, but not definitive for TA-1978).

iii. Mammalian cell mutagenesis (Gene point mutation induction).

Identification: DTX-77-0034, 6/29/77

Laboratory: Micro. Assoc. Bethesda, Maryland

Substance Tested: DAC-2787, 97.8% pure.

Cell Lines: Chinese hamster cells (V-79) and BALB/3 T3 mouse fibroblasts.

Summary:

V-79 cells (without activation) were exposed to 0.3 ugm/ml DAC-2787 for two hours. BALB/3T3 cells (with and without activation) were exposed to 0.3 ugm/ml for two hours. All cells were then plated and grown in the presence of ouabain 1 mM for 14 and 15 days respectively. [Positive control without activation was nitro-N-nitrosoguanidine (MNNG) and positive control with activation was benzo-(a)-pyrene]. Colonies were then counted.

Results:

DAC-2787 did not induce mutations at the ouabain-resistant locus in the inactivated V-79 cells nor in the BALB/3T3 cells with and without activation.

Conclusions:

Under the test conditions daconil is not an in vitro mutagen in V-79 hamster cells or in BALB/3T3 mouse fibroblasts.

iv. Ames bacterial mutagenicity assay

Identification: DTX-77-0035; 6/29/77

Laboratory: Micro. Assoc. Bethesda, Maryland

Substance test: DAC-2787, 97.8% pure

Cell Lines: TA-1535; TA-100; TA-1537 and TA-1538 (histidine-requiring) strains of ST.

Summary:

DAC-2787 was plated at 6.6, 3.3, 1.0, 0.66, and 0.33 ugm/plate against all strains with and without activation. DMSO (solvent) served as the negative control. MNNG, 2-nitroflourene (2-NF) and 9-aminoacridine (9-AA) served as unactivated positive controls and Aflatoxin B, and 6-aminochrysene (6-AC) served as activated positive controls. Plates were incubated and mutant colonies were counted.

Results:

DAC-2787 did not induce point mutations in any of the testor strains used, either with activation or without activation.

Conclusions:

DAC-2787 is not a bacterial mutagen in the Ames assay (to cell lines evaluated), at levels up to 6.6 ugm/plate.

v. DNA Repair/Ames Plate Assay

Study Identification: DTX-61-0002 (undated)

Laboratory: Inst. of Environmental Toxicology (Japan)

Substance Tested: DAC-2787. 99.3% pure

Cell Lines: DNA repair: B. subtilis, H-17 (wild) and M-44 (repair deficient). Ames assay: S. typh. (5 histidine dependent strains), E. Coli (2 strains).

Summary:

DNA repair strains were exposed to Daconil concentrations of 2, 5, 10. 20, 100 and 200 ugm/disk. Kanamycin served as negative control and mitomycin C served as positive control.

The Ames assay used Daconil concentrations of 1, 5, 10, 100 and 500 ugm/plate without activation, and concentrations of 2, 10 and 100 ugm/plate with activation.

DMSO was used as the negative control. 2-AA and 2-Af were positive controls.

Results:

Daconil failed to inhibit zones of B. subtilis and did not interfere with the repair mechanism of B. subtilis #M44. Daconil failed to increase the number of mutant colonies in any Ames testor strain.

Conclusions:

Under the conditions of this test Daconil is non-mutagenic and does not interfere with DNA repair mechanisms in selected bacterial strains.

vi. Ames Salmonella/microsomal assay

Study Identification: DTX-77-0038 6/29/77.

Laboratory: Microb. Assoc., Bethesda, Maryland.

Substance Tested: DS-3701 (4-OH metabolite), 99% pure.

Cell Lines: S. Typh. TA-1535, TA-100, TA-1538, TA-1537 and TA-98.

Summary:

DS-3701 was plated at levels of 100, 33.3, 10, 3.3 and 1 ugm/plate. Positive control substrates with activation contained MNNG, 2-NF and 9-AA. Positive controls without activation were 2-AA, aflatoxin B. and 6-AC. DMSO served as negative vehicle control. Plates were incubated at 37°C for 48 hours and evaluated.

Results:

DS-3701 failed to induce point mutations in any testor strain either in the presence of activation or in the absence of activation.

Conclusions:

DS-3701 is not an inducer of point mutations in selected strains of bacteria.

vii. DNA Repair Assay

Study Identification: DTX-77-0039, 6/29/77.

Laboratory: Microb. Assoc., Bethesda, Maryland

Substance Tested: DS-3701, 99% pure.

Cell Lines: S. Typh. TA-1538 (repair deficient) and TA-1978 (repair competent).

Summary:

DS-3701 was tested at 20, 10 or 2 ugm/plate with and without microsomal activation Non-activated positive controls were 6-AC and NQN activated control was 6-AC. Chloramphenicol served as negative control. Plates were incubated and evaluated for mutagenic events.

Results:

DNA was not affected by DC-3701. The zones of inhibition for each strain were similar.

Conclusions:

DNA repair mechanisms of S. typh. strains were not affected at plate levels up to 20 ugm DS-3701 plate.

viii. In vitro Mammalian Cell Point Mutation Assay

Study Identification: DTX-77-0040; 6/29/77.

Laboratory: Micro. Assoc., Bethedsa, Maryland.

Substance Tested: DS-3701, 99% pure.

Cell Lines: Chinesa Hamster cells (V-69) and mouse fibroblasts (BALB/3T3).

Summary:

Hamster and mouse cells were exposed for two hours to a growth medium containing 30 ug/ml of DS-3701. Cells were then seeded into flasks and incubated for 48 hours (hamster cells) or 96 hours (mouse cells). Following incubation, cells were plated into dishes containing 1mM ouabain and incubated for another 14 and 25 days, respectively. Ouabain resistant mutant colonies were then counted. MNNG served as the unactivated positive control. Acetone served as the negative control with activation. S-9 fraction alone, and heat inactivated S-9 plus BP was negative control without activation.

Results:

DS-3701 did not induce increased mutational frequency in either cell line, nor did it induce ouabain-resistant mutation either with or withour activation.

Conclusions:

DS-3701 is not mutagenic for hamster and mouse fibroblast cells at a concentration 1 mM/ml.

b. Metabolism Studies

i. Absorption, Excretion and Distribution of radio-labeled DAC-3701 in Rats.

Study Identification: R-78-00061; 8/18/78.

Laboratory: Diamond Shamrock Co., Painesville, OH.

Substance Tested: DAC-3701 with Spec. Act. of 4026 DPM/ugm., or 97% pure DAC-3701.

Animals - Four Sprague-Dawley males per dose.

Doses - Single PO 4.3 or 62.4 mg/kg in PEG-400.

Methods:

Material was gavaged into animals and urine and feces were collected and weighed daily for 96 hours. Food and water were available ad libitum. Rats were then killed and the lungs, stomach, gut, heart, kidney, liver, spleen and testes were reserved. Fat, blood, muscle and bone marrow were sampled. Cages were rinsed and the water was analyzed for radioactivity. All specimens were prepared for liquid scintillation spectroscopy. They were counted for two minutes each, except those requiring combustion and indirect $^{14}CO_2$ trapping which were counted for four minutes.

Recoveries of urine and feces were reported in terms of cummulative excretion of dose given initially. Post-mortem tissue recoveries were reported in terms of percent of total activity recovered. No attempt was made to characterize the metabolite(s) per se and all measurements were expressed in terms of ^{14}C content only.

Results:

Urine and Feces: ^{14}C excreted in the urine and feces were, as percentage of the dose, similar for the low-dose and the high-dose level animals. At the time of final collection (96 hours), urine and feces accounted for about 78% of the administered dose with 70% being found in the feces and 8% in the urine.

Tissues: Blood, muscle, gut and fat contained the bulk of the remaining activity about 15%; the gut had the greatest retention on a wet tissue weight basis.

Conclusions:

Seventy percent of the administered dose of DAC-3701 was removed via the GI tract. The remainder was found in the urine and in various tissues, notably the gut, blood and fat. The findings suggest that most of the DAC 3701 passes through the GI tract or is eliminated in the urine, but one cannot rule out the possibility that the daconil metabolite could be absorbed by the gut, metabolized in the liver and then excreted back into the gut via the bile.

c. Acute Toxicity Studies

i. Rabbit Eye Irritation Study

Study Identification: DTX-77-069; 11/22/77.

Laboratory: International Research and Development Corp. (IRDC).

Substance Tested: Technical DAC-2787, 96% pure

Animals: New Zealand White (NZW) Rabbits

Dose: 100 mg

Vehicle: None

Methods:

The material was instilled into the right eye of each of three males and three female NZW rabbits; the left eye served as control. Eyes were examined at 0, 24, 48 and 72 hours, and on days 7 and 14 using flourescein and UV transillumination. Lesions were graded after the method of Draize (1959)1/.

Results:

24 Hours: All treated eyes showed severe chemosis that precluded iridial and corneal exam.

48 Hours: All eyes showed corneal opacity, iridial and conjunctival irritation, which persisted through 72 hours.

72 Hours: Maximum irritation score of 60.8/110; opacity and iridial and conjunctival irritation.

7 Days: All eyes showing corneal opacity and irritation of iris and conjunctivae. Score of 49.2/110.

14 Days: 5/6 eyes showing corneal opacity. Average score 24.1/110.

Conclusions:

DAC-2787 is severe eye irritant.

CORE Rating: Guideline

TOX Catergory: I

Signal Word: Danger

ii. Rabbit Eye Irritation Study

Study Identification: DTX-77-0121; 1/25/78.

Laboratory: Springborn Institute for Bioresearch Inc. (SIBI). Spencerville, OH.

Substance Tested: Daconil 2787 Fungicide, 96% pure.

Animals - NZW rabbits

Dose - 100 mg

Vehicle - None

Methods:

The test substance was instilled into the right eye of each of three male and three females. The left eye served as control. Eyes were examined with flourescein and UV transillumination on days 0, 3 and 7. Eyes were examined without using the special technique at 24 and 48 hours and on day 14. Lesions were scored after Draize (1959).

Results:

A maximum score of 92.7/110 was observed on day 4. Injuries included corneal opacity, iris effects and conjunctival inflammation and discharge. The lesions were present in 5/6 rabbits at day 14, although they were decreased in intensity.

Conclusions:

DAC 2787 is a severe irritant in the eye of the rabbit.

CORE Rating: Guideline

TOX Category: I

Signal Word: DANGER

iii. Rabbit Eye Irritation Study

Study Identification: DTX-77-0125 2/14/78.

Laboratory: SIBI, Spencerville, OH

Substance Tested: Bravo W-75 (95% pure)

Animals - NZW rabbits

Dose - 100 mg

Vehicle - None

Methods:

Test substance was instilled into the right eye of each of three males and three females. The left eye served as controls. Eyes were examined using florscein on days 0, 1, 2, 3, 7 and 14. Lesions were scored after Draize (1959).

Results:

The test substance induced severe injuries in the exposed organs. A Draize score of 110/110 was reported. Deleterious effects were corneal opacity, iris effects, and conjuenctival swelling, inflammation and discharge. These responses had only minimally abated by day 14.

Conclusions:

Bravo 75W is a severe corrosive and irritating agent in the eye of the rabbit.

CORE Rating: Guideline

TOX Category: I

Signal Word: DANGER

iv. Acute Oral Toxicity in the Rat

Study Identification: DTX-78-0001; 6/30/78.

Laboratory: Bio-Dynamics Inc., E. Millstone, N.J.

Substance Tested: Daconil 2787

Animals - Sprague Dawley Rats

Vehicle - Tap water

Summary:

Groups of five male and five female adults were gavaged with 10.0, 14.1, 20 or 28.2 Gm/kg/bw. Body weights were taken initially and at termination on day fourteen. Animals were observed daily and mortality was recorded. Gross necropsy was performed at termination.

Results:

Signs of Toxicity: ataxia, rales, soft stools, urinary discoloration and lethargy.

Mortality:

3/10 rats died in the 20 Gm group and 2/10 died in the 28.2 and 10.0 gm groups. One of 10 at 14.1 gm/kg died.

Conclusions:

The oral LD_{50} in rats in this study is greater than 28.2 Gm/kg.

CORE Rating: Guideline

TOX Category: III

Signal Word: CAUTION

v. Acute Inhalation Study

Study Identification: DTX-78-0019

Laboratory: Bio-Dynamics Inc., Millstone, N.J.

Substance Tested: Daconil 2787 W-75; 2/14/79

Vehicle - Dry Air

Animals - Sprague-Dawley rats

Methods:

Groups of five female and five males were exposed for 4 hours to nominal aerial concentrations of test substance of 2.91, 1.38, 1.01, 0.69, 0.47 or 0.13 mg/L in a glass chamber whose dimensions were such as to contain 32.2 L. Animals were observed until death or until 14 days had passed. Terminal necropsies were done.

Results:

The four hour LC_{50} was determined to be 0.54 mg/L air. Signs of toxicity included apnea, lacrimation and possible hypothermia.

CORE Rating: Guideline

TOX category: II

Signal Word: Warning

vi. Rabbit Acute Dermal LD_{50}

Study Identification: DTX-78-0002; 5/12/78

Laboratory: Bio-Dynamics Inc., E. Millstone, N.J.

Substance Tested: Daconil 2787 Fungicide

Vehicle - Physiological Saline (slurry of 1 gm/ml)

Animals - NZW Rabbits

Methods:

An area of the lateral dorsi comprising about 30% of the total body area was clipped free of pellage in three male and three female adult animals and test substance was applied at a rate of 20 gm active/kg. The application site was occludel with impervious wrapping and Elizabethan collars were tied to the neck to prevent ingestion of material and removal of wrapping by the animals. After 24 hours the collars and occlusive wrappings were removed; and the area cleansed and evaluated for injury according to Draize (1944)2/. Animals were observed at 0, 2, 4 and 6 hours and daily thereafter for 14 days. Body weights were obtained initially and on days 7 and 14. Animals were killed and autopsied on day fourteen.

Results:

There was no mortality associated with the test substance; some dermal edema and erythema was noted.

Conclusions:

Daconil 2787 possesses a Dermal LD_{50} of greater than 20 gm/kg.

CORE Rating: Minimum

vii. Rat Acute Oral LD_{50}

Study Identification: DTX-78-0005; 6/30/78.

Laboratory: Bio-Dynamics Inc., E. Millstone, N.J.

Substance Tested: Bravo 75 W.

Vehicle - Tap Water

Animals - Sprague Dawley rats

Methods:

Five male and five female adult rats per dose level were gavaged with 7.1, 10.0, 14.1, 20.0 or 28.2 gm/kg. The animals were collectively housed and were observed twice daily for mortality and toxic signs for fourteen days. Animals were autopsied upon death or at termination.

Results:

Signs of toxicity included ataxia, urinary and fecal staining, prostration and lethargy. A dose-related increase in mortality was observed. The incidence was: 0/10, 2/10, 1/10, 6/10 and 8/10.

Conclusions:

The calculated Oral LD_{50} is 19.0 gm/kg in the rat.

CORE Rating: Minimum Data

Signal Word: CAUTION

TOX Category: III

viii. Rat Acute Inhalation Study

Study Identification: DTX-78-0008; 2/14/79

Laboratory: Bio-Dynamics Inc., E. Millstone, N.J.

Substance Tested: Bravo 75 W

Vehicle - Air

Animals - Sprague-Dawley rats

Methods

5 males and five females per group were exposed to nominal aerial concentrations of 0, 0.11, 0.45, 1.40, 2.88, or 18.3 mg/L air of test substance (These are values reported in the petitioner's summary; however, the report by the contracting laboratory show these values for nominal concentration to be 2.91, 1.38, 1.01, 0.69, 0.47 and 0.13 mg/L air respectively.) Calculations based on reported time intervals and amount of material delivered show that these values are correct except that the 2.91 value should be x 1.57 mg/L air), for four hours. Animals were placed in the exposure chamber and observed for health effects initially and at 15 minute intervals during the first hour, then each hour thereafter. Hourly observations were made for 4 hours post-exposure and daily for 14 days thereafter. Body weights were obtained initially and on days 1 (except for the 0.13 mg/L group), 2, 4, 7 and 14. Animals were killed and autopsied on day 14. The lungs and trachea from each animal were removed and prepared for possible histopathological examinatior.

Results:

Signs of Toxicity:

Lacrimation, nasal discharge, salivation, ano-genital staining, rales, dyspnea and "coldness".

Mortality:

All rats in the three high-dose groups perished; 6/10 in the 0.47 group died; 5/10 in the 0.69 group died 0/10 in the 0.13 mg/L groups.

Autopsy Findings:

Discoloration of major organs; presence of white particulate material in the tracheae and pulmonary irritation.

Conclusions:

Petitioner concluded that the LC_{50} in this study was 0.90 mg per liter; the contracting laboratory calculated 0.54 mg/L. However, since the 2.91 mg/L figure was incorrect, the value must be recalculated.

We are unable to explain why petitioner and the contracting laboratory have conflicting values.

CORE Rating: Supplementary

Repairable to: Guideline

How? 1. Resolve differences in exposure values between petitioner and contractor.

2. Recalculate the LC_{50}.

ix. Rabbit Acute Dermal LD_{50}

Study Identification: DTX-78-0006; 5/12/78.

Laboratory: Bio-Dynamics, Inc., E. Millstone, N.J.

Substance Tested: Bravo W-75

Vehicle - physiological saline

Animals - NZW Rabbits

Methods:

3 males and 3 female adult animals were prepared by clipping the pellage closely over the dorso-lateral surfaces, exposing an area approximately equal to 30% of the total body surface area. Test substance was applied as a 1 gm/ml slurry at the rate of 20 gm/kg body weight. The test site was occluded with impervious covering. Elizabethan collars were applied. After 24 hours of exposure the appurtenances were removed and the excess test substance was removed. Animals were scored for edema, erythema and eschar formation after the method of Draize (1944). Observations were made for mortality at 0 - 2 hours, at 4 - 6 hours after release; then once daily for fourteen days. Autopsies were performed on all animals dying during the study or at termination.

Results:

Moderate to severe erythema were noted at 24 hours. One male died on day 10, exhibiting clear nasal discharge and soft stool. All other animals showed clear nasal discharge.

Conclsions:

The tentative Dermal LD_{50} for this material is greater than 20 gm/kg.

CORE Rating: Minimum

Repairable: to: Guideline

How? Provide total skin area which the material was actually in contact with.

x. Rabbit Primary Dermal Irritation Study

Study Identification: DTX-78-0007; 5/12/78

Laboratory: Bio-Dynamics, Inc., E. Millstone, N.J.

Substance Tested: Bravo 75-W

Vehicle - water

Animals - NZW Rabbits

Methods:

Three male and three female rabbits were prepared by clipping closely the dorso-lateral pellage over an area equal to about 30% of the total body surface area. Test substance was applied as a 1 gm/ml slurry to each of two 1 in. x 1 in. sites, one abraded, one intact. 0.5 ml of test substance was covered with gauze and occluded with plastic and tape. 24 hours later the coverings were removed and the test sites were cleansed and evaluated after the method of Draize (1944) for erythrma, edema and eschar formation. Observations were made daily for seven days.

Results:

Slight to severe erythema and edema were noted through day five; two animals and some irritation up to day seven. Abrasion did not affect the response. The dermal irritation index was 2.0/8.0.

Conclusions:

Bravo 75-W is a moderate dermal irritant in the rabbit.

CORE Rating: Minimum data

TOX Category: II

4. Hexachlorobenzene (HCB)[3]/

Soybeans

Residues of HCB, a potential oncogen and contaminant of technical CTN, were shown to occur at levels no greater than 0.006 ppm from the proposed use. (RCB memo of P. Erricco, 8/13/80, PP#6F1799).

Parsnips

Residues of HCB in parsnips would be less that 0.04 ppm in parsnips (root portion) (memo of P. Errico, 2/20/80, PP#7E1887).

Citrus and Citrus Oil

No estimates are yet available from RCB as to HCB levels expected to occur in these rac; however additional information being requested of petitioner for this rac (See the RCB Review L. Bradley 1/30/81).

Overall, we conclude that, for the purpose of these tolerances, risk associated with potential residues of HCB in subject racs will parallel that of the CTN and its 4-OH metabolite; that is, exposure and therefore oncogenic risk is insignificant when compared to the theoretical risk attendant upon previously established tolerances for residue of CTN and its metabolite (including that for HCB).

REFERENCES

1. Draize, J.H., et al. Assoc. of Food and Drug Officials of U.S. Austin Texas, 1959. Ed. A. Lehman.

2. Draize, J.H., et al. J. Pharm. Expt. Therap. 82:337 (1944).

3. An in depth review of the toxicity of HCB including an oncogenic Risk Analysis has been prepared in draft form by Toxicology Branch, Roger Gardner, 12/8/80.

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