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

December 21, 2000

MEMORANDUM:

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

Subject: 032501: 1- and 3-day dermal toxicity study in rats 1% granular disulfoton (MRID# 45239602)(Non-Guideline study).

DP Barcode: D269999
Submission: S587191

From: David G Anderson
RRB-2 HED (7509C)

To: Christina Scheltema
SRRD (7508)

Thru: Alan Nielsen, BSS
RRB-2, HED (7509C)

The registrant submitted data on a non-guideline 1- and 3-day dermal study in rats of the 1% granular disulfoton. The DER for the study is attached. The reference and Executive Summary is below.

CITATION: Croutch, CR and Sheets, LP (2000). Repeat-Exposure (3-Day) Dermal Toxicity Study with 1% G Di-Syston®) in Rats. Testing Laboratory name Bayer Corp., Stilwell, KA. Laboratory report number: 109956, Study#00-S22-BS. October 16, 2000. MRID# 45239602. Unpublished

SPONSOR: Bayer Corp., Stilwell KA

EXECUTIVE SUMMARY: In a 3-day dermal rat study (MRID# 45239602) disulfoton, granular, 1% a.i. (1% G Di-Syston®)) was administered dermally to 5 Wistar (Crl:WI(HAN)BR) rats/sex/dose at 0, 50, 100, 200 or 500 mg/kg/day (equivalent to 0, 0.5, 1.0, 2.0 or 5.0 mg a.i./kg/day). Plasma and erythrocyte cholinesterase was measured at 24 hours after the first and day 3 dose. Brain cholinesterase was measured at termination on day 4. Test material was ground and applied to plastic backed gauze, moistened with water, applied to the shave the test site (about 10% of the body surface), then secured with a bandage. The animals were exposed dermally for 6 hour per day with washing at the end of the exposure period.

No clinical signs were noted or body weight decrement. No other signs of toxicity were noted, but the study was designed to determine cholinestersase depression only. After 1 day of dosing, the NOAEL in males was 200 mg/kg and the LOAEL was 500 mg/kg based on biologically significant 31% erythrocyte cholinesterase inhibition which was not statistically significant. After 1 day of dosing the NOAEL in females was 100 mg/kg and the LOAEL was

200 mg/kg based on biologically significantly increased inhibition of plasma cholinesterase (36%). After 3-days of dermal dosing the NOAEL in males was 100 mg/kg/day and LOAEL was 200 mg/kg/day based on a increase in brain cholinesterase inhibition of 21% (statistically significant). After 3-day of dosing the NOAEL in females was 50 mg/kg/day and the LOAEL was 100 mg/kg/day based on statistically significant plasma and brain cholinesterase inhibition of 37% and 18%, respectively.

The overall NOAEL of 100 mg/kg/day (equivant to 1.0 mg a.i./kg) with a LOAEL of 200 mg/kg/day (equivalent to 2.0 mg a.i./kg) based female plasma cholinesterase deprssion for 1 day of dosing. After 3 days of dosing the NOAEL was 50 mg/kg/day (equivalent to 0.50 mg a.i./kg/day) with a LOAEL of 100 mg/kg/day (equivalent to 1.0 mg a.i./kg/day) based on depressed plasma and brain cholinesterase in females.

The study is acceptable for a (NG) 1-day or 3-day dermal study in the rat.

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

Reviewer: David G Anderson, PhD,
Reregistration Branch-2 (7509C)
EPA Secondary Reviewer: Robert Fricke, PhD,
Reregistration Branch-2 (7509C)

Date ___

Date ___

DATA EVALUATION RECORD

STUDY TYPE: Cholinesterase Inhibition in a 3-Day Dermal Rat Study (NG)

DP BARCODE: D269999.
P.C. CODE: 0032501

SUBMISSION CODE:S587191
TOX. CHEM. NO.:455.

TEST MATERIAL (PURITY): [Disulfoton, 1% granular, purity: 1.0% a.i. (Systemic Rose and Flower Care®)]

SYNONYMS: *[O,O-Diethyl-S-[2--(ethylthio)ethyl]phosphorodithioate]*

CITATION: Croutch, CR and Sheets, LP (2000). Repeat-Exposure (3-Day) Dermal Toxicity Study with 1% G Di-Syston®) in Rats. Testing Laboratory name Bayer Corp., Stilwell, KA. Laboratory report number: 109956, Study#00-S22-BS. October 16, 2000. MRID# 45239602. Unpublished

SPONSOR: Bayer Corp., Stilwell KA

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No clinical signs were noted or body weight decrement. No other signs of toxicity were noted, but the study was designed to determine cholinestersase depression only. After 1 day of dosing, the NOAEL in males was 200 mg/kg and the LOAEL was 500 mg/kg based on biologically significant 31% erythrocyte cholinesterase inhibition which was not statistically significant. After 1 day of dosing the NOAEL in females was 100 mg/kg and the LOAEL was 200 mg/kg based on biologically significantly increased inhibition of plasma cholinesterase (36%). After 3-days of dermal dosing the NOAEL in males was 100 mg/kg/day and LOAEL was 200 mg/kg/day based on a increase in brain cholinesterase inhibition of 21% (statistically significant). After 3-day of dosing the NOAEL in females was 50 mg/kg/day and the LOAEL was 100 mg/kg/day based on statistically significant plasma and brain cholinesterase inhibition of 37% and 18%, respectively.

The overall NOAEL of 100 mg/kg/day (equivant to 1.0 mg a.i./kg) with a LOAEL of 200 mg/kg/day (equivalent to 2.0 mg a.i./kg) based female plasma cholinesterase deprssion for 1 day of dosing. After 3 days of dosing the NOAEL was 50 mg/kg/day (equivalent to 0.50 mg a.i./kg/day) with a LOAEL of 100 mg/kg/day (equivalent to 1.0 mg a.i./kg/day) based on depressed plasma and brain cholinesterase in females.

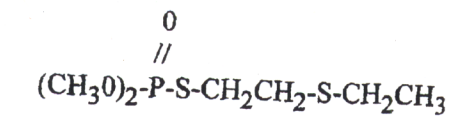
The study is acceptable for a (NG) 1-day or 3-day dermal study in the rat.

COMPLIANCE: Signed and dated GLP, Quality Assurance, Data Confidentiality, and Flagging statements were provided.

I. MATERIALS AND METHODS

A. MATERIALS

1. Test Material: 1% G Di-Syston®, approximately 1% a.i.
   Description: Granular (dark gray)
   Lot/Batch #: 041003/0030353
   Purity: 0.99% a.i.
   CAS #: 298-04-4
   Structure:



$(CH_30)_2$-P-S-CH_2CH_2-S-CH_2CH_3

2. Vehicle: water

3. Test animals: Rats: Wistar, strain. Age at start of dosing: 9 wks
   Weight at initiation males 235–247g and females174 to 180 g
   Source: Charles River Labs
   Housing: Suspended stainless steel
   Diet: Purina Mills Rodent Lab Chow, 5001-4; ad libitum
   Water: Municipal water supply ad libitum
   Environmental conditions:
   Temperature:19-25°C
   Humidity: 30%-70%
   Air changes: Not reported
   Photoperiod:12 hrs dark/12 hrs light

B. PROCEDURES AND STUDY DESIGN

1. Animals were asigned to 5 groups/sex ramdomly.
2. Animal assignment: Animals were randomly assigned by weight to test groups as seen in Table 1.

TABLE 1: Animal Assignment

Test Group	Dermal Dose in *(mg a.i./kg)*	Animals/group	
		Males	Females
0 mg/kg	0.0	5	5
50 mg/kg	0.5	5	5
100 mg/kg	1.0	5	5
200 mg/kg	2.0	5	5
500 mg/kg	5.0	5	5

3. Dose Administration: The backs of the test animals were shaved (about 10% of the body surface area). Granular (1%) Di-Syston® was pulvarized and weighed on to a two ply gauze with a plastic backing, moistened with water and applied to the test site and wrapped with a bandage (Vetrap) for 6 hour per day. The test area was wiped clean after the application each period. Doses were administered for three days.

4. Dose selection rationale: Dose selection was based on two 21-day rabbit dermal studies.

5. Dosage preparation and analysis: Since doses were used as measured, no dose analysis was necessary.

C. OBSERVATIONS

1. Animals: Daily observations and the schedule for those observations are summarized from the report.

2. Cholinesterase: Cholinesterase was determined by a modified Ellmans. 6,6'-dithioiodinicotinic acid was used as coupling agent to avoid hemoglobin interference rather than 5,5'-dithiobis-2-nitrobenzoic acid. The lab zeroed against a reagent blank for erythrocyte cholinesterase activity, and a tissue blank was substracted.

3. Blood was drawn for cholinesterase determination 24 hours after day 1 and day 3. Animals were not fasted prior to drawing blood.

4. Since cholinesterase levels were the only determinations required, histopathological examination was considered adequately conducted in previous studies in the rabbit.

B. RESULTS:

1. Clinical Observations: No clinical signs were seen.

2. Body Weight: Body weight and weight gain was unaffected.

3. Cholinesterase Measurements: All LOAELs were based on dose related decreases in cholinesterase and or increases in inhibition of cholinesterase. Plasma and erythrocyte cholinesterase measurements were conducted 24 hours after the first dose and 24 hours after the third dose. Brain cholinesterase measurement were conducted on whole brain homgenate at termination.

Table 1 gives the results form the plasma and erythrocyte cholinesterase determinations for males and females 24 hours after the first dose. After 1 day of dosing, the NOAEL in males was 200 mg/kg and the LOAEL was 500 mg/kg based on biologically significant 31% erythrocyte cholinesterase inhibition which was not statistically significant. After 1 day of dosing the NOAEL in females was 100 mg/kg and the LOAEL was 200 mg/kg based on biologically significantly increased inhibition of plasma cholinesterase (36%).

Table 2 gives the results from plasma, erythrocyte and brain cholinesterase determinations in males and females 24 hours after 3-days of dermal dosing. After 3-days of dosing the NOAEL in males was 100 mg/kg/day and LOAEL was 200 mg/kg/day based on a increase in brain cholinesterase inhibition of 21% (statistically significant). After 3-day of dosing the NOAEL in females was 50 mg/kg/day and the LOAEL was 100 mg/kg/day based on statistically significant plasma and brain cholinesterase inhibition of 37% and 18%, respectively.

Table 1: Plasma and erythrocyte cholinesterase levels in males and females 24 hours after the first dose.

Dose of 1% granular disulfoton	Plasma Che IU/mL±SD	% inhib.	Eryth. Che IU/mL±SD	% inhib.
		Males		
0 mg/kg	0.54±0.07	-	1.13±0.40	-
50 mg/kg	0.51±0.06	6	1.18±0.29	0
100 mg/kg	0.48±0.04	11	1.13±0.30	0
200 mg/kg	0.52±0.05	4	0.99±0.36	12
500 mg/kg	0.46±0.05	15	***0.78±0.26***	***31***
		Females		
0 mg/kg	1.85±0.69	-	0.97±0.12	-
50 mg/kg	1.64±0.42	11	0.94±0.24	3
100 mg/kg	1.58±0.49	15	1.26±0.16	0
200 mg/kg	***1.18±0.36***	***36***	0.91±0.25	6
500 mg/kg	***0.62±0.14***	***67****	***0.65±0.11***	***33****

* = p≥0.5; Bolded/italics are considered different from control

Table 2: Plasma, erythrocyte and brain cholinesterase levels in males and females 24 hours after the third dose.

Dose	Plasma Che IU/mL±SD	% inhib. in plasma	Eryth. Che IU/mL±SD	% inhib. erythrocytes	Brain Che IU/ml±SD	% inhib. in brain
			Males			
0 mg/kg	0.48±0.06	-	0.86±0.21	-	13.2±0.8	-
50 mg/kg	0.47±0.09	2	0.88±0.11	0	12.4±1.0	6
100 mg/kg	0.46±0.07	4	0.98±0.27	0	12.6±0.4	5
200 mg/kg	0.46±0.06	4	0.68±0.21	21	***10.4±0.7****	***21****
500 mg/kg	***0.31±0.09****	***35****	***0.50±0.37***	***42****	***9.0±1.1***	***32****
			Females			
0 mg/kg	1.63±0.50	-	1.09±0.29	-	13.6±0.4	-
50 mg/kg	1.27±0.27	22	0.66±0.21	39*	12.4±1.0	9
100 mg/kg	***1.03±0.18****	***37****	0.95±0.29	13	***11.2±0.7****	***18****
200 mg/kg	***0.53±0.11***	***68****	***0.91±0.25***	***62****	***8.1±1.1****	***40****
500 mg/kg	***0.35±0.14***	***79****	***0.30±0.17***	***72****	***3.6±0.7****	***74****

* = p≥0.5; Bolded/italics are considered different from control.

C. STUDY DEFICIENCIES: The only study deficiency that could be seen was that the study was conducted only for 3-days, when it was uncertain that a 3 day study would be adequate to characterize residential/non-occupational exposure.