3-13-84

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

003680

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

TO: William Miller, PM#16
Registration Division (TS-767)

FROM: Amal M. Mahfouz, PhD
Toxicologist, Section V
Toxicology Branch/HED (TS-769)

TRHU: Laurence D. Chitlik, DABT
Section Head, Section V
Toxicology Branch/HED (TS-769)
and
THRU: William L. Burnam, Chief
Toxicology Branch
Hazard Evaluation Division (TS-769)

SUBJECT: PP#4F2996. Terbufos; (0,0-diethyl phosphorodithioate), EPA Reg.#241-238. A Petition for Permanent Tolerances of Terbufos and its ChE-Inhibiting Metabolites in/on Peanut Shells at 2.50 ppm and in/on Peanut Nutmeat at 0.05 ppm Using Counter 15G Formulation.
Accession Nos.: 072149 and 072150; CASWELL#131A

Petitioner: American Cyanamid Company
Agricultural Research Division
P.O. Box 400
Princeton, New Jersey 08540

Chemical: 0,0-diethyl S-(1,1-dimethylethyl methyl) phosphorodithioate

Synonyms: Counter, Terbufos, CL 92 100, AC 92 100

Action Requested:

1. American Cyanamid requested the following permanent tolerances for Terbufos in/on peanuts using Counter 15G formulation at planting and at pegging:

Peanut shells: 2.50 ppm
Peanut nutmeat: 0.05 ppm

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2. The following new mutagenicity studies were submitted for review with this petition:

1. Chromosome Aberration in Chinese Hamster Ovary Cells
2. CHO/HGPRT Mutation Assay
3. Rat Hepatocyte Primary Culture/DNA Repair Test

3. The registrant also indicated that the recently submitted (9/7/83) histopathological reevaluation of the 24-month oral toxicity and carcinogenicity studies in rats should be considered as a part of this petition (in order to obtain a higher priority review). However, these histopathological data have been already reviewed in my 12/28/83 memo to W. Miller (RD).

Recommendation:

A. The requested permanent tolerances for Terbufos in/on peanuts cannot be toxicologically supported at the present time due to the existing extensive data gaps for this chemical. These data gaps are as follows:

°Technical Material

- Acute Dermal LD_{50} in Rabbits (F).
- Acute Inhalation LC_{50} in Rats (M&F).
- Two Teratology Studies in two different animal species; rat and rabbit are perferred.
- Two-Year Chronic Feeding Study in Rats.
- One-Year Feeding Study in Dogs.
- Lifetime Oncogenic Study in Mice.
- Additional Mutagenicity Studies:
  1. In vivo cytogenetics test for chromosomal aberrations using bone marrow preparations of rats.
  2. Dominant lethal test in rats or mice.

°Counter 15G Formulation

- Acute Oral LD_{50} in Rat (F).
- Acute Dermal LD_{50} in Rabbit (F).

B. The following 3 mutagenicity studies submitted with this petition were found negative and were classified as acceptable:

1. Chromosome aberration in Chinese Hamster Ovary Cells.
2. CHO/HGPRT Mutation Assay.
3. Rat Hepatocyte Primary Culture/DNA Repair Test.

However, since the above studies were negative, two additional studies are required. These 2 studies are listed above in section A under data gaps for mutagenicity.

C. All the previously submitted chronic feeding studies (in rats and dogs) provide only supplementary data and cannot be used at the present time as bases for tolerance assessment. However tolerances for terbufos use have been already established on racs under CFR 40, 180.352 (See the attached computer printout).

The incremental dietary exposure from food uses has been assessed for the proposed new use in/on peanut nutmeat as follows:

- Existing TMRC for published and unpublished tolerances = 0.0046 mg/day (1.5 kg diet)
- Increase in TMRC due to the new use = 0.0002 mg/day/1.5 kg diet
- Percent increase in the TMRC due to the proposed new use = 4.5

D. Counter 15G formulation is acutely toxic [LD_{50} for dermal exposure in rabbits = 10.2 (7.7-13.4) mg/kg, Toxicity Category I]; thus, it should be classified in the restricted use Category. Due to this high toxicity, I also recommend that the label should clearly indicate that the applied chemical (banded) should be incorporated in the soil.

E. The Terbufos Registration Standard should be amended to reflect the present status of data gaps as stated in section A above and to indicate the successful filling of 3/5 data gaps for mutagenicity as indicated in section B above.

Residue Chemistry Considerations:

This petition has not yet been reviewed by the Residue Chemistry Branch.

Review

A. Petition Review

1. Formulation:

The formulation proposed for use on peanuts is Counter 15G which is 15% (by weight) terbufos. The inerts are cleared under Section 180.1001.

2. Directions for Use

See the attached copy of the supplemental labeling.

3. Referenced Petitions:

PP#3G1340 - 0.05 ppm - corn grain, fodder & forage.
PP#4F1496 - 0.05 ppm - corn grain, fodder & forage.
PP#5F1640 - 0.05 ppm - in or on sugar beets tops & roots
PP#6F1657 - 0.5 ppm - corn forage and fodder (field corn, popcorn and sweet corn), and 0.05 ppm - corn grain (popcorn) and sweet corn (kernels plus cob with husks removed).
PP#1F2433 - 0.05 ppm - cabbage, broccoli and cauliflower
PP#1F2540 - 0.05 ppm be established for sorghum grain and 0.5 ppm be established for sorghum forage and fodder for combined residues of the insecticide.
PP#2F2608 - 0.05 ppm - soybean grain
PP#3F2926 - 0.05 ppm - rape and mustard seed

However tolerances were only established for sugar beets, corn and sorghum, see CFR 40 (1983), section 180.352; and the attached computer printout.

4. Three new mutagenicity studies were submitted with this petition and they are reviewed in the following section B.

5. Previously submitted toxicity data are summarized and attached to this memo.

6. Data gaps for this chemical are extensive; they are summarized in the Recommendation section (A), page 2.

7. PADI Calculation. The provisionary acceptable daily intake (PADI) for Terbufos was based on a 6-month dog feeding study (FDRL #1193, 12/6/72), using a NOEL for ChE inhibition of 0.10 ppm (0.0025 mg/kg/day) and a 100-fold safety factor. However, this study was recently downgraded to supplementary in the Terbufos Registration Standard, 12/30/82.

Hence, a new ADI/PADI will be calculated as soon as a new chronic feeding study in rats or a one-year feeding study in dogs is available.

Terbufos toxicology reviews

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Pages _____ through _____ are not included in this copy.

The material not included contains the following type of information:

___ Identity of product inert ingredients

___ Identity of product impurities

___ Description of the product manufacturing process

___ Description of product quality control procedures

___ Identity of the source of product ingredients

___ Sales or other commercial/financial information

X A draft product label

___ The product confidential statement of formula

___ Information about a pending registration action

___ FIFRA registration data

___ The document is a duplicate of page(s) _____

___ The document is not responsive to the request

The information not included is generally considered confidential by product registrants. If you have any questions, please contact the individual who prepared the response to your request.

CFR 180.352

1/24/84

File last updated: 3/23/83

ACCEPTABLE DAILY INTAKE DATA

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Dog mg/kg	NOEL ppm	S.F.	ADI mg/kg/day	MPI mg/day(60kg)
~~0.003~~	~~0.10~~	~~100~~	~~0.0000~~	~~0.0015~~
0.33	9.9	100	0.099	5.94

Published Tolerances

CROP	Tolerance	Food Factor	mg/day(1.5kg)
Sugar,cane&beet(154)	0.150	3.64	0.00273
Corn,grain(68)	0.050	1.50	0.00075
Corn,sweet(40)	0.050	1.43	0.00107
Sorghum(147)	0.050	0.03	0.00002

MPI	TMRC	% ADI
0.0015 mg/day(60kg)	0.0046 mg/day(1.5kg)	~~304.91~~ 304.93

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Current Action 4F2996

CROP	Tolerance	Food Factor	mg/day(1.5kg)
Peanuts(115)	0.050	0.36	0.0027

MPI	TMRC	% ADI
0.0015 mg/day(60kg)	0.0048 mg/day(1.5kg)	~~322.80~~ 324.3

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DRAFT

8. RPAR status. The compound is not on the RPAR list and there are no pending regulatory actions against this chemical to this reviewer's knowledge.

B. Review of the New Data

1. Chromosome Aberrations in Chinese Hamster Ovary Cells (CHO); AC 92,100.

Accession Number: 072149

Sponsor: American Cyanamid Company, Princeton, N.J. 08540
Study #981-83-106

Testing Facility: Microbiological Associates, 1530 East Jefferson St, Rockville, Md. 20852. Study #T.1906.337006.

Dates: From 2/21/83 to 5/5/83; report dated 5/10/83; study submitted to EPA on 11/22/83.

Study Director: Dr. A. Thilager of Microbiological Associates.

Test Material: AC 92,100, lot #AC-4391-1, a viscous liquid stored at room temperature. Upon my request on 1/5/84, the registrant indicated on 1/6/84 (orally to the product manager) that the test article was the technical product with 87.8% a.i.

Procedure: The test compound was initially examined in duplicate for cytotoxicity determinations. Based on this test, six decreasing dose levels from 100 nl/ml were used to evaluate the in-vitro chromosomal aberrations in the chinese hamster ovary cells. The dosages tested were 100, 50, 25, 10, 5 and 2.5 nl/ml. Each dose level was tested with and without microsomal activation. Rat liver microsomal enzyme preparations, S-9, from Arochlor-induced rats were used for activation. The test dosages were dissolved in DMSO 10-20 minutes before it was added in triplicates to culture flasks seeded with 5×10^5 cells/flask. The cultures were prepared 24 hours prior to treatments.

The cells were exposed for 2 hours to the test compound with activation; then they were washed twice with phosphate buffer saline (PBS), refed with fresh medium and allowed an expression period of 12-14 hours before harvest. In the non-activation assay, the cells were exposed to the terbufos dosages for a total period of 12 to 14 hours.

Examinations for chromosomal aberrations were performed in two of each triplicate tests after treatment with 0.1 ug/ml of colcemid in order to arrest the dividing cells at metaphase. The third flask was used to determine the mitotic index per dose level.

Triethylenemelamine (TEM) was used as the positive control for the non-activated system, and cyclophosphamide (CP) was the positive control for the activated system.

The cells were scored for chromosome aberrations and mitotic index.

Results:

All cells treated at 100 nl/ml dosage level died in both tests with and without activation. Cells treated at the 50 nl/ml level and above in tests without activation had a zero mitotic index.

The results of this in-vitro chromosomal assay with CHO were negative in both the absence and presence of S-9 as demonstrated in table #1 on next page.

Discussions:

This reviewer notes that: 1) Only one harvest of cells was examined for chromosomal aberrations instead of 3 harvests. However, the registrant did not provide any information concerning the cell cycle at the dosages tested or the proportions of the population of cells in various stages of the cell cycle at these dosages. Hence, the negative mutagenic potential reflected by these data cannot be adequately assessed in this study alone without additional mutagenicity data and/or studies; 2) Except for the lowest dose examined, the mitotic index at each of the higher dosage levels was generally lower than the index for the negative control and for the solvent control in both tests with and without activation. Also the 3 highest dosages examined for chromosomal aberrations in tests without activation reflected a lower mitotic index than the positive control (see table #1 on next page). The significance of these findings remain to be explained.

Conclusions:

This study reflects a negative mutagenic potential for terbufos and may be considered as conditionally acceptable. However, the registrant should address the issues discussed in the above section concerning the number of harvests per cell culture and the mitotic index before this study can be considered as fully acceptable.

Study Classification: Acceptable (however, see the above section concerning the need for additional information).

Table #1. Data on Mitotic Activities.

Without Activation			With Activation		
Test Article Concentration	Mitotic[1] Index	Relative[2] Mitotic Index	Test Article Concentration	Mitotic Index	Relative Mitotic Index
50 nl/ml	0	0	50 nl/ml	2.4	40.68
25 nl/ml	2.1	35.59	25 nl/ml	4.1	69.49
10 nl/ml	2.5	42.37	10 nl/ml	5.4	91.53
5 nl/ml	3.7	62.71	5 nl/ml	7.3	123.73
2.5 nl/ml	6.0	101.69	_____	___	_____
Positive Control TEM 1.0 ug/ml	2.8	45.16	Positive Control CP 50.0 ug/ml	1.9	22.09
Solvent Control	5.9	100.00	Solvent Control	5.9	100.00
Negative Control	6.2	100.00	Negative Control	8.6	100.00

[1]Mitotic Index = Average number of dividing cells/100 cells examined ; note that the total no. of cells examined was 1000 cells per dosage (100 cells per count, 10 counts per slide)

[2]Relative Mitotic Index = $\frac{\text{Mitotic Index of test dose}}{\text{Mitotic Index of Solvent Control}} \times 100$

2. In Vitro Mammalian Cell Mutation (HGPRT) in Chinese Hamster Cells (CHO); AC 92,100.

Accession No.: 072149

Sponsor: American Cyanamid Company
Princeton, New York 08540

Testing Facility: Same as Sponsor.

Study No.: CHO-4602 and 4603; Project #0402

Dates: From 5/14/83 to 8/16/83; report dated 10/21/83; submitted to EPA on 11/22/83.

Study Performed By: E. Johnson and B. Fine, and the study was reported by J. Allen and E. Johnson.

Test Material: AC 92,100; batch #AC 4391-1, 87.8% a.i.; a liquid.

Vehicle: Dimethylsulfoxide (DMSO).

Positive Controls: Methane sulfonic acid, ethyl ester (EMS) for tests without activation (at 200 ug/ml); and Benz (a) anthracene, 7,12-dimethyl (7,12 DMBA) for tests with activation (at 7 ug/ml).

Cell Line: CHO-K_1-BH_4 cells were obtained from J.P. O'Neill, Oak Ridge National Laboratory.

Procedure: A range finding cytotoxicity test was initially performed using 7 terbufos concentrations from 10 to 1000 ug/ml. Based upon these data the highest dosage selected for CHO/HGPRT mutation assay was 100 ug/ml. The dosages used in this point mutation assay were 100, 75, 50, 25 and 10 ug/ml. Arochlor-induced rat liver enzyme preparation, S-9, was used in the tests with activation. Solvent control (0.1 ml DMSO), and the appropriate positive control (as mentioned above under the positive controls section) were used in both tests with and without S-9 activation. Note that the 7,12-DMBA test without S-9 activation was considered as a negative control in this study; however this reviewer did not use it for comparisons because values from this control group appeared to be slightly lower than the solvent control values.

Due to the reaction of the test material with the plastic culture dishes at the high dosage levels, both glass and plastic (only at the lower dosages) dishes were used in the cytotoxicity section of this study. It is not clear if glass dishes were used in the remaining tests.

The cells were initially seeded in 25 cm^2 flask and washed 3 times with phosphate buffered saline (PBS) after an incubation period of 24 hours; then they were treated with the test material. The cells were exposed for 5 hours to the assigned dosages, then washed with PBS and incubated overnight before additional testings.

For the mutagenicity evaluations, the above cells were subcultured on day 1 and at 2 day-intervals up to a period of 9 days after exposure. For each subculture, 10^6 cells were used. On day 9, the subcultured cells were plated for determination of the mutation frequencies. Aliquots of 2 x 10^5 cells/100 mm dishes (using a hypoxanthine free medium which contained 10 uM 6-thioguanine) were used in these determinations, allowing the examination of 10^6 cells per dose (5 dishes). These tests were run in triplicate contingent upon the presence of a sufficient number of cells. An additional 200 cells from the final subculture was plated in hypothanthine free medium for evaluation of the cloning efficiency of these cells.

Also 200 cells from day 1 subculture were used for cytotoxicity determinations.

Results:

1. The initial cytotoxicity tests indicated that terbufos is highly toxic to CHO cells (0% survival after 5 hrs exposure) in glass dishes at 100 ug/ml dose level and above (200, 300, 500 and 1,000 ug/ml) in tests with activation; however, in tests without activation, high toxicity (0% survival) was also noted at 50 ug/ml and above.

The cells survivability was much higher at the lower dosages (100, 50 and 10 ug/ml) with and without activation when the cells were tested in plastic dishes, (except for the 10 ug/ml dose level with activation, the lowest dose tested, it reflected a lower survivability, 91.7% as compared to the cells tested in glass dishes, 109.4%).

Cloning efficiency was also parallel to cytotoxicity.

2. In the mutagenicity tests, cytotoxicity was dose-related as summarized below:

Dosages (ug/ml)	100	75	50	25	10	0*	Positive Control
Average % Survival**:							
With S-9	1.2	12.4	32.4	52.4	73.5	100.0	48.2
Without S-9	4.5	4.0	15.1	32.4	61.8	100.0	33.2

*Vehicle (solvent) control, 0.1 ml DMSO.
**Average counts of five dishes containing 2 x 10^2 cells each in triplicate per dose. The average % survival was also considered as the cloning efficiency.

In the above table, a high level of cytotoxicity was noted at the 100 and 75 ug/ml dose levels. This cytotoxicity limited the number of cells available for the evaluation of the mutagenic frequencies at these dosages (only one determination was made the 75 ug/ml level in tests with and without activation; and the 100 ug/ml dosage was not examined for mutagenic frequencies).

3. The mutagenic frequencies were examined in triplicates for the 50, 25 and 10 ug/ml dosage levels. The mutagenic frequencies at the dosage levels were similar to data from the solvent control in tests without activation; also in these tests, the value determined for 75 ug/ml dosage was within the range of the solvent control values.

In tests with S-9, the mutagenic frequencies noted at the lowest dose tested were similar to the control values; however, two of the triplicate used at 25 and 50 ug/ml reflected a slightly higher level of mutation. The 75 ug/ml dosage reflected a much higher level of mutation frequencies than the solvent-control and the lower dosage levels. However, the value obtained at the 75 ug/ml dosage level was 3x to 6x lower than the positive control group, see the table below:

Dosage in ug/ml	Mutation Frequencies X 10^{-6} Without S-9	With S-9
75	2.9	< 50
50	< 1.8 to 2.2	3.5 to 16
25	< 2.2 to 5.3	< 2.5 to 16.8
10	< 1.2 to 4.6	2.6 to 2.2
0*	3 to 12.1	< 1.5 to 2.8
Positive Control**	175 to 304.1	157.9 to 305.2

*Solvent control.
**The positive control for the test without S-9 was EMS; for tests with S-9 it was 7,12 DMBA.

Discussions:

As noted above, tests without S-9 clearly gave negative results. However (as also noted by the registrant), statistical analysis of the data obtained from the tests with S-9 reflected effects at 25 and 50 ug/ml dosages when these data were compared to the study solvent control. The registrant also stated that no significant difference was noted when these data were compared to historical solvent control data (the historical data were not included in the report); however, comparisons with the solvent control data for tests without S-9 (see above table) did not reflect a significant difference. For clarification of these findings, the registrant repeated the tests with S-9. The following dosages were used 5, 10, 25, 33, 42 and 50 ug/ml and the results were clearly negative when compared (by inspection or statistically) to the new solvent data.

Conclusions: The study is acceptable and reflects a negative mutagenic potential for terbufos.

Study Classification: Acceptable

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3. Rat Hepatocyte Primary Culture/DNA Repair Test (HPC/UDS); AC 92,100.

Accession No.: 072149

Sponsor: American Cyanamid Company
Princeton, New York 08540

Testing Facility: Pharmakon Research International, Inc. Waverly, Pennsylvania 18471. Study #PH 311-AC-001-83.

Dates: From 3/9/83 to 5/4/83; report dated 5/5/83; submitted to EPA on 11/22/83.

Study Director: Edmund G. Bodek of Pharmakon.

Test Material: AC 92,100; batch #AC 4391-1, 87.8% a.i.; a liquid.

Vehicle: Dimethylsulfoxide (DMSO)

Test System: Liver of one male Fischer 344 rat. The liver was perfused, excised and combed, yielding 7.78 x 10^5 cells per ml was 260 gm in weight and was obtained from the Charles River Laboratories.

Dosages: 0.33, 1.0, 3.33, 10.0, 33.33, 100.0, 333.33, 1000.0, 3333.3 and 10,000.0 ug/well in 2 ml of media.

Positive Control: 2-Acetoamidofluorene (2AAF) was used at a concentration of 1 x 10^{-4} M.

Procedure: The male rat was anesthetized (intraperitoneally) with 50 mg/kg sodium pentobarbital. The animal was open and the liver was perfused with Medium A* then Medium B* at a constant temperature, 37°C. The liver was then excised, placed in a sterile culture dish with 50 ml of median B and combed with a camel's hair brush. The cells were pelleted and their viability was measured with trypan blue dye exclusion. An aliquot of 5 x 10^5 viable hepatocytes was cultured for 2 hours at 37°C then washed and tested in a serum-free medium containing the test compound and 10 uC/ml tritiated thymidine (specific

*"Medium A - 0.5 mM ethylene-glycol-bis-(B-aminoethyl ether)-N-N' - tetraacetic acid (EGTA) in Ca^{++} -Mg^{++} free Hank's balanced salt solution buffered with 10 mM N-2-hydroxyethyl-piperazine-N'-2-ethanesulfonic acid (Hepes), pH adjusted to 7.35, sterilized through a 0.2 u filter and maintained at 37°C.

**Medium B - Collagenase [Sigma Type I], 100 Units/ml prepared in serumless Williams' Medium E (WME) (GIBCO), buffered with 10 mM Hepes, pH adjusted to 7.35. Gentamicin was added to the final volume of 0.05 mg/ml. This perfusion media was sterilized through a 0.45 u filter unit and placed in a 37°C water bath before and during perfusion."

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activity 50-80 uC/mM). The cells were exposed to the test substance and H-thymidine for 20 hours, then washed with phosphate buffered saline, swelled in 1% sodium citrate, fixed and mounted on glass slides. The slides were dropped in NTB-2 photographic emulsion, dried overnight and stored at 4°C for 1 week. The slides were then washed, stained, covered and examined for unscheduled DNA repair synthesis.

Cytotoxicity was noted at 100 ug/well and above. Hence, the highest dose used for determination of DNA repair test was 33.33 ug/well and below, these dosages were tested in triplicate. A positive control, a vehicle control and an untreated control were used for the comparison of data.

Unscheduled DNA repair synthesis values were quantified as the net increase in black silver grains over the nucleus for 50 cells/slides as compared to values from the highest of three cytoplasmic grain counts of areas (approximately equal the nucleus size) adjacent to the nucleus. The slides were read at 1500x magnification. These data were recorded in this study report as average counts per triplicate per dose.

Results:

Terbufos reflected a negative potential for mutagenicity at 33.33 ug/well and below in the rat hepatocyte primary culture/DNA repair test. The next higher dosage, tested 100 ug/well, and above were cytotoxic. Table 1 below reflect these data:

"Table 1. Autoradiographic Analysis of DNA Repair in the Rat Hepatocyte Primary Culture/DNA Repair Test

Treatment	Concentration/ml of Media	Net Nuclear Grains Triplicates Cultures $\bar{x}$ s.d.
Untreated		0.3 ± 0.3
DMSO		0.2 ± 0.1
2AAF	1×10^{-4}M	49.7 ± 1.1**
AC 92,100	0.33 ug/well	0.4 ± 0.5
AC 92,100	1.0 ug/well	1.2 ± 0.8
AC 92,100	3.33 ug/well	1.4 ± 1.2
AC 92,100	10.00 ug/well	0.3 ± 0.4
AC 92,100	33.33 ug/well	1.0 ± 0.8
AC 92,100	100.00 ug/well	Cytotoxic
AC 92,100	333.33 ug/well	Cytotoxic
AC 92,100	1000.00 ug/well	Cytotoxic
AC 92,100	3333.33 ug/well	Cytotoxic
AC 92,100	10,000.00 ug/well	Cytotoxic

**Positive finding. Mean net nuclear grain count of five or greater than the vehicle control."

Discussions:

Although the above study appears to be acceptable, this reviewer notes the following fact that may limit the usefulness of these data:

1. The standard deviation for the values reported for terbufos treated hepatocytes is unacceptably large, i.e. in this study, the standard deviations were often equal and sometimes higher than the mean (see table #1 on page 12 of this review).

2. The reliability of the automatic counter may be limited (as discussed with I. Mauer and W. Schneider on 2/27/84).

3. This chemical is acutely toxic in live animals (LD_{50} in rats is 1.3 mg/kg) and appears to be highly cytotoxic with a narrow toxic range. As noted in table #1, no dosage was tested between the cytotoxic dose of 100 ug/well and the non-cytotoxic dose of 33.33 ug/well. Hence, in view of the toxic nature of this chemical, it is not clear from this study if this test was performed at adequate dosage levels.

4. A liver from only one male animal was tested.

Conclusions: This study appears to be acceptable and is clearly negative within the dosage levels tested in this study when compared to the positive control. However, see my concerns as listed in the discussion section above.

Study Classification: Acceptable

I reiterate that this study is acceptable within the context of the discussions in the above section and in consideration of the fact that the additional battery of mutagenic studies are still required (see recommendation #B on page 3 of this memo paragraph #2).