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

APR - 9 2001

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

MEMORANDUM

Subject: Additions/corrections to [MRID 449753-04] Subchronic Oral Toxicity Study [870.3100 (§82-1)] with Immunotoxicity [870.7800 (§85-5)], Neurotoxicity [870.6200 (§83-1, §82-7, §81-8)] and One-Generation Reproduction in the Rat with Glycolic Acid

EPA Reg. No.:071654-R
Glycolic Acid
DP BARCODE: D264010, D264011, D264012, D264014
SUBMISSION CODE: S576708
P.C. CODE: 000101
EPA ID. NO.: 071654-R

From: S. L. Malish, Ph.D., Toxicologist, RASSB/AD S. L. Malish 3/21/01

To: Robert Brennis, PM 32
PM Team Reviewer, Marianne Clark
Regulatory Management Branch
Antimicrobials Division (7510C)

Thru: Winston Dang, Team Leader, Team One,
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (AD)[7510C]

3/21/2001

and

Norman Cook, Chief, RASSB/AD (7510C) 04/09/01

Applicant: E.I. Dupont de Nemours, Inc., Wilmington, DE

Action: Additions/corrections to "One-Generation Reproduction in the Rat" with Glycolic Acid section of MRID 449753-04 and the accompanying memorandum.

FORMULATION:

Active Ingredient:	% by weight
Glycolic Acid	≈70%

Laboratory: E.I. Dupont de Nemours, Inc.'s Haskell Laboratories for Toxicology and Industrial Medicine, Newark, DE

I. One-Generation Reproduction in the Rat with Glycolic Acid

The following paragraph should replace the last paragraph on page 4 of the **Memorandum [Toxicity Review** of Subchronic Oral Toxicity Study [870.3100 (§82-1)] with Immunotoxicity [870.7800 (§85-5)], Neurotoxicity [870.6200 (§83-1, §82-7, §81-8)] and One-Generation Reproduction in the Rat with Glycolic Acid] and the last paragraph on p. 32 of MRID 449753-04, entitled **Toxicity Review** of Subchronic Oral Toxicity Study [870.3100 (§82-1)] with Immunotoxicity.....].

This study is considered **Acceptable/Non-guideline**. The study does not fulfill the guideline [OPPTS 870.3800 (§83-4)] for a reproduction study in the rat. [note: a two generation guideline study would have included ≈30 animals /sex/group, a F_1 mating, P_1 reproductive organ weights with gross and histopathology and in the F_1 animals that were selected for mating, reproductive organ weights with gross and histopathology].

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

2/01/2001

OFFICE OF
PREVENTION, PESTICIDES AND
TOXIC SUBSTANCES

FEB - 1 2001

MEMORANDUM

Subject: Toxicity Review of Three Mutagenicity Studies
EPA Reg. No.:071654-R
Glycolic Acid
DP Barcode: D264011, D264012, D264014
Case: 062360
PC Code: 00101

From: S. L. Malish, Ph.D., Toxicologist, RASSB/AD S.L. Malish 1/23/01

To: Robert Brennis, PM 32
PM Team Reviewer, Marianne Clark
Regulatory Management Branch
Antimicrobials Division (7510C)

Thru: Winston Dang, Team Leader, Team One, 1/26/2001
Risk Assessment and Science Support Branch (RASSB)
Antimicrobials Division (AD) [7510C]

and

Norman Cook, Chief, RASSB/AD (7510C) Norman J. Cook 02/5/2001

Applicant: E.I. Dupont de Nemours, Inc., Wilmington, DE

FORMULATION:

Active Ingredient:	% by weight
Glycolic Acid	≈70%

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Laboratory: E.I. Dupont de Nemours, Inc.'s Haskell Laboratories for Toxicology and Industrial Medicine, Newark, DE

ACTION REQUESTED: Review the following toxicity studies (MRID 449753-05,449753-06, 449753-07) for mutagenic potential of the test compound, Glycolic Acid.

CONCLUSIONS: Glycolic Acid was not considered to be a mutagen, in study 1 and 2 of the mutagenicity studies listed below. In study 3, [mouse lymphoma cell assay], a mutagenic response was seen only at dose concentrations 3 to 6X higher than the Agency's maximum concentration (10 mM) for this type of study. **Glycolic acid was, therefore, not considered a mutagen for, regulatory purposes,** in this study.

All studies are considered **Acceptable** by the reviewer and fulfill the guideline requirements for these types of studies.

Executive Summaries:

The executive summaries are listed below. Complete data evaluation documents are attached.

1. *Salmonella typhimurium* and *Escherichia coli* Reverse Mutation Assays (MRID 449753-05)

In a reverse gene mutation assay (MRID 44975305) in bacteria, *S. typhimurium* strains TA97a, TA98, TA100 and TA1535 and *E. coli* strain WP2uvrA (pKM101) were exposed to Glycolic acid (70.58% a.i., lot number not provided) at concentrations of 1, 5, 10, 50, 100, 500, 1000, 2500 and 5000 µg/plate in the presence and absence of mammalian metabolic activation (S9-mix).

Concentrations of 1000 µg/plate and higher generally caused a reduction in the number of revertants per plate compared to solvent control values and a thinning or absence of the background lawn of bacteria in all strains, with and without S9-mix.

There was no increase in the number of revertants per plate in any strain over solvent control values at any concentration of Glycolic acid tested, with or without S9-mix. The solvent and positive controls induced the appropriate responses in the corresponding strains. **There was no evidence of induced mutant colonies over background.**

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This study is classified as **Acceptable** and satisfies the requirement for FIFRA Test Guideline OPPTS 870.5265 and 870.5100 (§ 84-2) for *in vitro* mutagenicity (bacterial reverse gene mutation) data.

2. *In Vivo* Mammalian Cytogenetics: Erythrocyte Micronucleus Assay in Mice (MRID 449753-07)

In a Crl:CD-1 (ICR)BR mouse bone marrow micronucleus assay (MRID 449753-07), five mice/sex/dose/harvest time were treated once each via oral gavage with Glycolic acid (Batch No. not provided, 70.58% a.i.) at doses of 300, 600 and 1200 mg/kg in males and concentrations of 400, 800 and 1600 mg/kg in females. Bone marrow cells were harvested at 24 hours post-treatment from all dose groups and also at 48 hours post-treatment from the 1200 [♂] and 1600 [♀] mg/kg dose groups.

There were signs of toxicity during the study, including lethargy, moribundity and/or abnormal gait appearing within two hours post-treatment in a few mice of both sexes. In addition, four males and two females from the high dose groups were found dead on the day following dosing and two additional mice, one male and one female, from the high dose groups were found dead two days post-treatment. No statistically significant (5% level) decreases in body weight gain were seen in any Glycolic acid treated group. There were no statistically significant increases in the frequency of micronucleated PCEs over solvent control values in either sex in any of the Glycolic acid treated groups. A slight decrease in the proportion of PCEs/total erythrocytes was seen in males and females at the 48 hour harvest time, possibly indicating some bone marrow toxicity; however, the decreases were not statistically significant. Positive and solvent control values were appropriate and within the testing laboratory's historical control ranges.

There was no significant increase in the frequency of micronucleated polychromatic erythrocytes in bone marrow at any dose or treatment time over background.

This study is classified as **Acceptable** and satisfies the requirement for FIFRA Test Guideline OPPTS 870.5395 (§ 84-2) for *in vivo* cytogenetic mutagenicity data.

3. Mammalian Cells in Culture Gene Mutation Assay in L5178Y TK$^{+/-}$ Mouse Lymphoma Cells (MRID 449753-06)

In a mammalian cell gene mutation assay at the TK locus (MRID

44975306), L5178Y $TK^{+/-}$ mouse lymphoma cells cultured *in vitro* were exposed to Glycolic acid (Batch No. not provided, 70.58% a.i.) at concentrations of 39.3, 78.5, 157, 313, 625, 1250, 2500 and 5000 µg/mL in the absence of mammalian metabolic activation (S9-mix) and at concentrations of 250, 500, 1000, 2000, 2500, 3000, 4000 and 5000 µg/mL in the presence of S9-mix. The S9-fraction was obtained from Aroclor 1254 induced male Sprague-Dawley rat liver.

Glycolic acid was tested up to a limit concentration of 5000 µg/mL. No cytotoxicity was seen in a preliminary test with ten concentrations of Glycolic acid ranging from 9.85 to 5000 µg/mL, with and without S9-mix. Minimal cytotoxicity was seen both with and without S9-mix in two mutagenicity assays. The average relative growth of the solvent controls was approximately 99% with or without S9-mix in both assays. The average relative growth of Glycolic acid treated cultures was approximately 50% at 5000 µg/mL with or without S9-mix in both assays.

In the initial mutation assay, both with or without S9-mix, a positive response [two-fold or greater increase in mutant frequency compared to the solvent control value] was seen only at 5000 µg/mL. The mutant frequency showed a 4.6X increase with S9-mix and a 2.9X increase without S9-mix compared to the controls. The positive response seen in the absence of S9-mix in the initial assay was not reproduced in the confirmatory assay. However, the positive response seen with S9-mix in the initial assay was also seen in the confirmatory assay. A positive dose-response increase compared to the control was obtained in the 4 dose range of 2500 µg/mL (32.9 mM)[2.02X] through 5000 µg/mL (65.8 mM) [4.59X]. The mutant colonies were predominantly small colonies, indicating a clastogenic mechanism of action. Positive and solvent controls gave the appropriate response.

Although Glycolic acid was mutagenic in the presence of S9-mix as tested in this study, mutagenic activity was only seen at concentrations 3 to 6X above the maximum testing concentration recommended by the EPA guidelines for this assay (10 mM). **For regulatory purposes, therefore, Glycolic acid, was not considered to be a mutagen.**

This study is classified as **Acceptable** and satisfies the requirement for FIFRA Test Guideline, OPPTS 870.5300 (§ 84-2) for *in vitro* mammalian forward gene mutation data.

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