



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

008455

JUL 16 1991

OFFICE OF
PESTICIDES AND TOXIC SUBSTANCES

MEMORANDUM

SUBJECT: Trifluralin: Review of a dermal absorption study in monkeys

Caswell No. 889
MRID No. 406737-01

HED Project No. 1-1078
EPA ID #: 036101

TO: Terri Stowe, PM Team 71
Special Review and Registration Division (H7508W)

FROM: Whang Phang, Ph.D. *Whang 7/10/91*
Pharmacologist
Tox. Branch II/HED (H7509C)

THROUGH: James Rowe, Ph.D. *James V. Rowe 7/11/91*
Section Head
and
Marcia van Gemert, Ph.D. *J.M. Loannon for MVE. 7/11/91*
Branch Chief
Tox. Br. II/HED (H7509C)

A dermal absorption study in rhesus monkey was reviewed by Robert Zendzian, Ph.D.. A DER and the reviewer's memorandum to James Rowe, Ph.D. are attached. The conclusion is as follows:

Significant uncertainties make the actual dose per unit area impossible to determine, and, therefore, the percent absorbed values are meaningless. Further monkey studies should be discouraged. The study is unacceptable, and the data can not be used for risk assessment purposes.



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

008455

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

MEMORANDUM

June 26, 1991

SUBJECT: Trifluralin, Dermal Absorption Study in Monkeys

TO: James Rowe Ph.D.
Head, Review Section III
Toxicology Branch II, HED

FROM: Robert P. Zendzian Ph.D. 6/26/91
Senior Pharmacologist
SACB, HED (H7509C)

Action Requested

Review the following study;

Percutaneous (dermal) absorption of [14C]-Trifluralin in rhesus monkeys, E.R. Adams, S.J. Glass & R.B.L. van Lier, Toxicology Division, Lilly Research Laboratories, P07487 & P03087, May 11, 1988, MRID 406737-01

Conclusion

The study is Unacceptable.

Significant uncertainties make the actual dose, per unit area, impossible to determine and, therefore, the percent absorbed values are meaningless. Future monkey studies should be discouraged. The data cannot be used for risk assessment purposes.

Discussion

This study generally follows the Maibach protocol for dermal absorption in primates (monkey or man), in which the same individual is given a dermal dose and an intravenous dose of the test compound and the urinary excretion of the intravenous dose is used to correct the urinary excretion of the dermal dose in order to obtain the portion of dose absorbed by the dermal route. Using this correction it is not necessary to collect or account for the portions of the dose excreted by other routes. The correction is as follows;

$$\frac{\% \text{ Dermal Dose in Urine}}{\% \text{ Intravenous Dose in Urine}} = \% \text{ Dermal Dose Absorbed}$$

Table 3. Corrected absorbed doses. All values are percent of administered dose.

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
Urine dermal dose	0.04	0.09	0.05	0.04
Urine intravenous dose	63.59	53.37	66.62	64.33
Corrected dermal absorption	0.06	0.17	0.08	0.06

Considering the problems associated with a material ballance study in the monkey the recovery, except IV for animal 15461, is good. Excluding that animal, dermal absorption for a 24 hour exposure is no more than 0.1 % and 11% of the dermal dose is unaccounted for compared to the IV dose. However, as noted below the uncertianties in the study are such that these values cannot be supported and should not be used for risk assesment.

There are four problems associated with this study, 1) the dermal dose was mg/kg rather than mg/cm², 2) the actual dermal dose was not quantitated, 3) use of an acetone swab following the soap and water wash of the application site and 4) the large portion of the dose found on the stainless steel screen covering the application site.

1) The rate at which a dermal dose penetrates the skin is proportional to the dose in mass per unit area. Penetration increases with increasing dose per unit area but the increase is not directly proportional to the increase in dose. The animals in this study were dosed on a mg/kg basis with each individual dose applied to an equal skin area. This results in different mg/cm² doses for each animal. Table 4 presents the calculated dose per unit area for the animals in this study. The differences in absorption secondary to the differences in dose per unit area probably would not be detectable with trifluralin because of the small quantity apparently absorbed and the relative insensitivity of the experimental design but with another compound they could be significant.

Table 4. Dermal dose per unit area. A dose of 2 mg/kg was applied to an area of 6 cm².

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
weight, kg	3.8	4.2	5.5	6.3
dose, mg/animal	7.6	8.4	11.0	12.6
dose, mg/cm ²	1.27	1.40	1.83	2.10

2) Applying and spreading a dermal dose usually leaves part of the dose on the application device. In this study the dose "was applied evenly to a 6 square centimeter area on the forearm and the vehicle evaporated with a hair dryer." No mention was made as to what was used to apply the dose nor was the residue on the application device determined. Considering the small volume administered, 0.05 ml/kg, this residue could represent a significant portion of the nominal dose.

3) Swabbing the application site with acetone does not occur in the field and it removed 1.3 to 2.7 % of the dose that was potentially available for continued absorption.

4) The major problem associated with this study is that most of the dermal dose was found on the stainless steel screen covering the application site. From 51 to 64 percent of dose was not available for absorption and there is no way of knowing why or how rapidly this happened. The report states, "The relatively large fraction of the applied dose found on the application screen is consistent with the high volatility observed with trifluralin in other studies (Parochetti et al., 1976)."

Volatilization of the dose would not be expected to deposit the material on the screen and have it stay there. Vaporized trifluralin would be expected to continue off into the atmosphere and be lost to the test. Any material depositing on the screen would be purely transient. It is more likely that the material on the screen resulted from the screen rubbing on the skin and removing test material somewhat like sandpaper on a painted surface. This could have occurred any time during the 24 hour exposure period such that the actual dermal dose was considerably less than nominal.

Together the deficiencies are such that the study must be classified unacceptable. An acceptable single dose, single duration study could have been performed with four rats according to the Agency protocol.

Attachment DER

Compound TrifluralinCitation

Percutaneous (dermal) absorption of [14C]-Trifluralin in rhesus monkeys, E.R. Adams, S.J. Glass & R.B.L. van Lier, Toxicology Division, Lilly Research Laboratories, P07487 & P03087, May 11, 1988, MRID 406737-01

Reviewed by Robert P. Zendzian Ph.D.
Senior Pharmacologist

Core classification unacceptable

Conclusion

Significant uncertainties make the actual dose, per unit area, impossible to determine and, therefore, the percent absorbed values are meaningless. Monkey studies should be discouraged.

Materials

Trifluralin, EL-152, 326EF8, 100.1% pure

Trifluralin, EL-152, 553-KB0-216 (ringlabeled) 8.59 uCi/mg
99.6% radio pure

Rhesus monkeys, four adults, 2 males and 2 females

Experimental Design

Test animals were first dosed dermally and then, after 7 days, dosed intravenously, both doses with 2 mg/kg of trifluralin.

Dosing material

For the dermal study test material was dissolved in ethanol to give a final concentration of 40 mg/ml and a specific activity of 1.5 uCi/mg.

For the intravenous study test material was dissolved in ethanol to give a final concentration of 10 mg/ml and a specific activity of 1.5 uCi/mg.

Dosing, Dermal study

Animals were placed in metabolism chairs for the first 24 hours, and then placed in individual metabolism cages. In the chairs, animals were fixed so as to be unable to contact the application site.

"Twenty-four hours prior to dosing, the right ventral forearm of each monkey was shaved. Aliquots of the prepared dose solution were collected for determination of radiocarbon. A volume of 0.05 ml of [^{14}C]-trifluralin/kg body weight, equivalent to 2.0 mg/kg, was applied evenly to a 6 square centimeter area on the forearm and the vehicle evaporated with a hair dryer. The application site was covered with a stainless steel screen supported by a neoprene window. After twenty-four hours, the screen and support window were removed and the application site was washed with Ivory Dishwashing Soap and water. The wash water was saved and the volume measured. The application site was then swabbed with an acetone saturated gauze pad. The screen, wash water and gauze were all saved for determination of radiocarbon content."

Dosing, Intravenous study

"Each monkey was administered a dose of 0.2 ml of [^{14}C]-trifluralin/kg of body weight, equivalent to 2.0 mg/kg, injected into the saphenous vein." Animals were then placed individually in metabolism cages for 7 days.

Samples

"In each study, blood samples were collected from the femoral vein of each monkey immediately before dosing and at 0.25, 0.5, 1, 2, 4, 6, 24, 48, 72, 96, 120, 144 and 168 hours after dosing."

"In each study, total urine and feces samples were collected for 24 hours prior to dosing and at 6, 24, 30, 48, 72, 96, 120, 144 and 168 hours after dosing."

Results

Dermal study.

Recovery from the application site, plasma concentrations, urinary excretion and fecal excretion for each animal are presented in Tables 6, 3.2, 4.2 and 5.2 respectively from the report. The data are summarized in Table 1 below.

Table 1. Recovery, as percent of dose, after dermal dose. Data from Tables 6, 4.2 and 5.2 from the report.

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
Application site	82.3	78.5	80.3	75.8
Urine	0.04	0.09	0.05	0.04
Feces	0.05	0.00	0.01	0.03
Total	82.39	78.59	80.36	75.87

Intravenous study.

Plasma concentrations, urinary excretion and fecal excretion from the individual animals are presented in Tables 3.1, 4.1 and 5.1 respectively from the report. Recovery is summarized in Table 2 below.

Table 2. Recovery, as percent of dose, after intravenous dose. Data from Tables 4.1 and 5.1 from the report.

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
Urine	63.59	53.37	66.62	64.33
Feces	20.51	16.06	24.56	18.44
Total	84.10	69.43	91.18	82.77

Discussion

This study generally follows the Maibach protocol for dermal absorption in primates (monkey or man), in which the same individual is given a dermal dose and an intravenous dose of the test compound and the urinary excretion of the intravenous dose is used to correct the urinary excretion of the dermal dose in order to obtain the portion of dose absorbed by the dermal route. Using this correction it is not necessary to collect or account for the portions of the dose excreted by other routes. The correction is as follows;

$$\frac{\% \text{ Dermal Dose in Urine}}{\% \text{ Intravenous Dose in Urine}} = \% \text{ Dermal Dose Absorbed}$$

Table 3. Corrected absorbed doses. All values are percent of administered dose.

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
Urine dermal dose	0.04	0.09	0.05	0.04
Urine intravenous dose	63.59	53.37	66.62	64.33
Corrected dermal absorption	0.06	0.17	0.08	0.06

Considering the problems associated with a material balance study in the monkey the recovery, except IV for animal 15461, is good. Excluding that animal, dermal absorption for a 24 hour exposure is no more than 0.1 % and from 11% of the dermal dose is unaccounted for compared to the IV dose. However, as noted below the uncertainties in the study are such that these values cannot be supported and should not be used for risk assessment.

There are four problems associated with this study, 1) the dermal dose was mg/kg rather than mg/cm², 2) the actual dermal dose was not quantitated, 3) use of an acetone swab following the soap and water wash of the application site and 4) the large portion of the dose found on the stainless steel screen covering the application site.

1) The rate at which a dermal dose penetrates the skin is proportional to the dose in mass per unit area. Penetration increases with increasing dose per unit area but the increase is not directly proportional to the increase in dose. The animals in this study were dosed on a mg/kg basis with each individual dose applied to an equal skin area. This results in different mg/cm² doses for each animal. Table 4 presents the calculated dose per unit area for the animals in this study. The differences in absorption secondary to the differences in dose per unit area probably would not be detectable with trifluralin because of the small quantity apparently absorbed and the relative insensitivity of the experimental design but with another compound they could be significant.

Table 4. Dermal dose per unit area. A dose of 2 mg/kg was applied to an area of 6 cm².

<u>Animal number</u>	<u>16021</u>	<u>15461</u>	<u>10731</u>	<u>10721</u>
weight, kg	3.8	4.2	5.5	6.3
dose, mg/animal	7.6	8.4	11.0	12.6
dose, mg/cm ²	1.27	1.40	1.83	2.10

2) Applying and spreading a dermal dose usually leaves part of the dose on the application device. In this study the dose "was applied evenly to a 6 square centimeter area on the forearm and the vehicle evaporated with a hair dryer." No mention was made as to what was used to apply the dose nor was the residue on the application device determined. Considering the small volume administered, 0.05 ml/kg, this residue could represent a significant portion of the nominal dose.

3) Swabbing the application site with acetone does not occur in the field and it removed 1.3 to 2.7 % of the dose that was potentially available for continued absorption.

4) The major problem associated with this study is that most of the dermal dose was found on the stainless steel screen covering the application site. From 51 to 64 percent of dose was not available for absorption and there is no way of knowing why or how rapidly this happened. The report states, "The relatively large fraction of the applied dose found on the application screen is consistent with the high volatility observed with trifluralin in other studies (Parochetti et al., 1976)."

8

Volatilization of the dose would not be expected to deposit the material on the screen and have it stay there. Vaporized trifluralin would be expected to continue off into the atmosphere and be lost to the test. Any material depositing on the screen would be purely transient. It is more likely that the material on the screen resulted from the screen rubbing on the skin and removing test material somewhat like sandpaper on a painted surface. This could have occurred any time during the 24 hour exposure period such that the actual dermal dose was considerably less than nominal.

Together the deficiencies are such that the study must be classified unacceptable. An acceptable single dose, single duration study could have been performed with four rats according to the Agency protocol.

9

Page _____ is not included in this copy.

Pages 11 through 16 are not included.

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 quality control procedures.
 - ☐ Identity of the source of product ingredients.
 - ☐ Sales or other commercial/financial information.
 - ☐ 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.

Tox Chem No. trifluralin

File Last Updated

Current Date

EPA
MRID

Study/Lab/Study #/Date	Material	LD50, LC50, PIS, NOEL, LEL	TOX Category	CORE Grade/Doc. No.
Dermal Absorption, Monkey; Lilly; P07487, P03087; 5/11/88	[14C]trifluralin technical	406737-01	N/A	UNAcceptable
Results: Significant uncertainties make the actual dose, per unit area, impossible to determine and, therefore, the percent absorbed values are meaningless. Monkey studies should be discouraged.				