AD-A126 372 PRELIMINARY ASSESSMENT OF THE RELATIVE TOXICITY OF NN-DIPROPYLCYCLO-HEXAN..(U) ARMY ENVIRONMENTAL HYGIENE AGENCY ABERDEEN PROVING GROUND MD J A MACKO ET AL. 1/1

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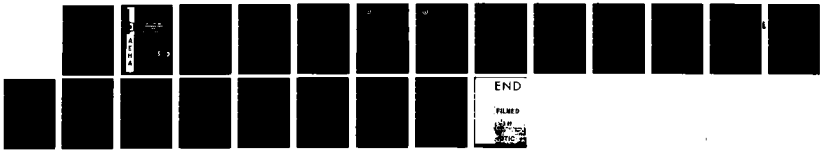

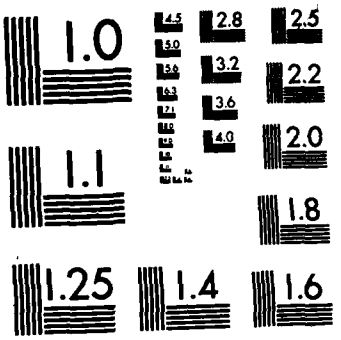

MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS-1963-A

UNITED STATES ARMY ENVIRONMENTAL HYGIENE AGENCY

ABERDEEN PROVING GROUND, MD 21010

AEHA

PHASE 1
PRELIMINARY ASSESSMENT OF THE RELATIVE TOXICITY
OF N,N-DIPROPYLCYCLOHEXANECARBOXAMIDE,
AI3-36326, IN LABORATORY ANIMALS
STUDY NO. 75-51-0233-83
NOVEMBER 1979 - AUGUST 1982

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REPORT DOCUMENTATION PAGE		READ INSTRUCTIONS BEFORE COMPLETING FORM
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4. TITLE (and Subtitle) Phase 1, Preliminary Assessment of the Relative Toxicity of N,N-Dipropylcyclohexanecarboxamide AI3-36326, in Laboratory Animals, Study No. 75-51-0233-83, November 1979-August 1982		5. TYPE OF REPORT & PERIOD COVERED Preliminary November 1979 - August 1982 6. PERFORMING ORG. REPORT NUMBER
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18. SUPPLEMENTARY NOTES

19. KEY WORDS (Continue on reverse side if necessary and identify by block number)

N,N-Dipropylcyclohexanecarboxamide	insect repellent
AI3-36326	irritation
acute	sensitization
dermal	mutagenicity
oral	enzyme induction

20. ABSTRACT (Continue on reverse side if necessary and identify by block number)

Technical grade N,N-Dipropylcyclohexanecarboxamide was not acutely toxic by ingestion or by dermal exposure, nor did it cause any phototopic or skin sensitization reaction. It causes slight to mild primary skin irritation and slight to mild reversible injury to the cornea and conjunctiva, both effects seeming dependent upon batch sample being tested. AI3-36326 induced enzyme production in rats with a subsequent decrease in hexobarbital sleeping time.

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20. (continued)

Care should be exercised when this compound is handled in the area of the eyes. Should ocular exposure occur, rinsing the eyes with water is effective in reducing corneal irritation. Variations in skin and ocular response to different batch samples of AI3-36326 indicate the need for chemical stabilization of this chemical before proceeding with large scale chronic tests.

Phase 1, Study No. 75-51-0233-83, Nov 79 - Aug 82

CONTENTS

DEPARTMENT OF THE ARMY
U. S. ARMY ENVIRONMENTAL HYGIENE AGENCY
ABERDEEN PROVING GROUND, MARYLAND 21010

Mr. Macko/jr/AUTOVON
584-3980

REPLY TO ATTENTION OF

HSHB-OT/WP

31 MAR 1983

SUBJECT: Phase 1, Preliminary Assessment of the Relative Toxicity of N,N-Dipropylcyclohexanecarboxamide, AI3-36326, in Laboratory Animals, Study No. 75-51-0233-83, November 1979 - August 1982

Executive Secretary
Armed Forces Pest Management Board
Forest Glen Section, WRAMC
Washington, DC 20307

EXECUTIVE SUMMARY

The purpose, essential findings and recommendations of the inclosed report follow:

a. Purpose. The purpose of these studies was to acquire information on the acute toxicity of N,N-dipropylcyclohexanecarboxamide (AI3-36326) in laboratory animals. Results from these studies will allow an assessment of the risk from short-term exposure to this compound and will provide guidance for further subchronic toxicological tests.

b. Essential Findings. Technical grade carboxamide was not acutely toxic by ingestion or by dermal exposure, nor did it cause any phototoxic or skin sensitization reactions. It caused mild primary skin irritation and slight to mild reversible injury to the cornea and conjunctiva, both effects seemingly dependent upon batch sample being tested. AI3-36326 induced enzyme production in rats with a subsequent decrease in hexobarbitol sleeping time.

c. Major Recommendations. Stablization and characterization of AI3-36326 should be accomplished before further toxicologic evaluation. Essential additional tests should include a standard 21-day rabbit dermal and a rat teratology study. Protocols should be developed to evaluate the potential microsomal enzyme induction mechanisms of AI3-36326 and a biological assay for skin irritation responses between different production lots. Individuals who work with this compound should exercise care to prevent eye splash exposure. To prevent irritation, in the event of exposure, rapid eye rinse with copious amounts of water should be performed.

FOR THE COMMANDER:

1 Incl
as

JOHN W. CUTTING, M.D.
Colonel, MC
Director, Occupational and
Environmental Health

CF:
HQDA (DASG-PSP) wo incl
Cdr, HSC (HSPA-P)
Comdt, AHS (HSHA-IPM)
Dir, Advisory Cen on Tox, NRC (2 cy)
USDA, ARS (Dr. Terrence McGovern)
USDA, ARS-Southern Region

DEPARTMENT OF THE ARMY
U. S. ARMY ENVIRONMENTAL HYGIENE AGENCY
ABERDEEN PROVING GROUND, MARYLAND 21010

REPLY TO ATTENTION OF

HSHB-OT/WP

PHASE 1
PRELIMINARY ASSESSMENT OF THE RELATIVE TOXICITY OF N,N-DIPROPYLCYCLOHEXANECARBOXAMIDE,*†
AI3-36326, IN LABORATORY ANIMALS
STUDY NO. 75-51-0233-83
NOVEMBER 1979 - AUGUST 1982

1. AUTHORITY.

a. Memorandum of Understanding between the US Army Environmental Hygiene Agency; the US Army Health Services Command; the Department of the Army, Office of The Surgeon General; the Armed Forces Pest Management Board; and the US Department of Agriculture, Agricultural Research, Science and Education Administration, titled, Coordination of Biological and Toxicological Testing of Pesticides, effective 23 January 1979.

b. Letter, AFPMB, Armed Forces Pest Control Board, Washington, DC, 29 November 1979, subject: Toxicological Testing of Candidate Repellent Compounds, with inclosure.

2. REFERENCES. See Appendix A for a listing of references.

3. PURPOSE. Studies were conducted to acquire information concerning the acute toxicity of N,N-dipropylcyclohexanecarboxamide, AI3-36326, in laboratory animals. Results from these studies will provide guidance for further toxicological testing of this compound.

4. BACKGROUND. A prime objective of the United States Department of Agriculture's repellent research program is to support the entomological needs and requirements of the US Army (paragraph 1a, this report). The chemical utilized in this study has shown potential as a substitute or replacement for the current standard insect repellent N,N-diethyltoluamide (M-DET). The proposed use of the candidate insect repellent would be the topical application to human skin as a liquid or spray, and/or for the treatment of clothing via spraying or total immersion. Treatment rate would be determined to provide maximum protection equivalent to or better than M-DET. The subject insect repellent AI3-36326 has been tested as 25 or 12.5 percent concentrates in ethanol (250 or 125 mg/forearm application). However, intended use concentrations might range between 6.25 percent and 50 percent in liquid cream formulations and/or aerosol sprays (paragraph 1b, this report).

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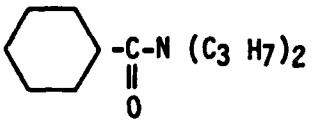
* In conducting the studies described in this report, the investigators adhered to the "Guide for the Care and Use of Laboratory Animals," US Department of Health, Education and Welfare Publication No. (NIH) 74-23, revised 1978, and reprinted in April 1980 as NIH Publication No. 80-23.
† The experiments reported herein were performed in animal facilities fully accredited by the American Association for the Accreditation of Laboratory Animal Care.

5. MATERIAL. Insect repellent AI3-36326, N,N-dipropylcyclohexanecarboxamide was synthesized by Terrence P. McGovern, Ph.D., Organic Chemical Synthesis Laboratory, Agricultural Research Center, US Department of Agriculture, Beltsville, MD 20705 (reference 1, Appendix A). It is a light tan liquid with a molecular weight of 209, a chemical formula of $C_{13}H_{25}NO$, and a slightly irritating odor. The batch sample "E" was analyzed upon receipt, using infrared (IR) spectrophotography (Appendix B) and gas chromatography (GC) and was found to be free of any significant impurities. The chemical structure of AI3-36326 is as follows:



6. PROCEDURE.

a. New Zealand White rabbits were used for skin and eye studies, Hartley guinea pigs for a sensitization study and Sprague-Dawley rats for determination of oral toxicity and enzyme induction studies.† Five strains of Salmonella typhimurium were used in evaluating the mutagenic potential of AI3-36326. Scoring for the evaluation of skin and eye reactions are shown in Appendices C and D, with explanation of USAEHA categories in Appendix E.

b. All animals were maintained on commercial chow ration¶ with water available ad libitum. Housing was in rooms with a 12-hour light-dark sequence under ambient conditions of 24°C ± 2°C and 40-45 percent relative humidity. Rabbits were housed in individual wire cages while rats and guinea pigs were housed in groups of five in hanging gang cages.

c. All tests were conducted following toxicology's Standing Operating Procedures (SOP) (references 2 and 3, Appendix A).

---

† Rabbits and guinea pigs were purchased from Dutchland Laboratories, Inc., Denver, Pennsylvania. The rats were selected from our breeding colony, offspring of Caesarian Originated, Barrier Sustained (COBS) rats from Charles River Breeding Laboratories Inc, Wilmington, Massachusetts.
¶ Certified Rodent Chow 5002, Rabbit Chow 5322 and Guinea Pig Chow 5026 from Ralston Purina Company, St Louis, Missouri.

Phase 1, Study No. 75-51-0233-83, Nov 79 - Aug 82

7. FINDINGS. A tabular presentation of toxicity data developed in this Agency follows:

TABLE. PRESENTATION OF DATA

Test	Results	Interpretation
SKIN IRRITATION STUDIES		
Rabbits		
Single 24-hour application to intact and abraded skin of New Zealand White rabbits. 0.5 mL of each batch sample, -c, -d and -e, technical grade compound applied to each of six rabbits.	Batch numbers -c, -d, and -e of compound AI3-36326 all caused slight to well defined irritation responses of intact and abraded skin.	AI3-36326-c USAEHA Category II. AI3-36326-d USAEHA Category III. AI3-36326-e USAEHA Category I.
EYE IRRITATION STUDIES		
Rabbits		
Single 24-hour application of 0.1 mL batch sample AI3-36326-c technical grade compound to one of each of six New Zealand White rabbits.	Batch -c caused mild irritation to the cornea and conjunctiva in all six rabbits.	AI3-36326-c USAEHA Category C.
Application made to additional rabbits of 0.1 mL propylene glycol and 10 and 1% propylene glycol solutions of AI3-36326-c.	Propylene glycol caused no irritation. 10% AI3-36326-c in propylene glycol caused moderate injury to the cornea and conjunctiva in all rabbits.	Propylene glycol USAEHA Category A. 10% AI3-36326-c USAEHA Category E.

Test	Results	Interpretation
	1% AI3-36326-c in propylene glycol caused no corneal changes and very slight conjunctival irritation.	1.0% AI3-36326-c USAEHA Category A.
Single 24-hour application of 0.1 mL batch sample AI3-36326-e technical grade compound to one eye of each of six New Zealand White rabbits. Three additional rabbits received 0.1 g chemical to one eye followed in 20 seconds by a 1 minute gentle wash with warm tap water.	Non-washed out eye showed 3 of 6 rabbits with small area of opacity completely clear at 72 hrs. No irritation in eyes washed with warm water.	AI3-36326-e USAEHA Category A batch -e much less irritating than other batches. Methods in chemical synthesis of compound may affect irritant responses. Washing eliminates possible irritant effects of technical grade compound.
PHOTOCHEMICAL SKIN IRRITATION STUDIES		
Test conducted according to Toxicology SOP, Basic Safety Assessment Test Procedure for Primary Dermal Photochemical Skin Irritation Study in Rabbits, November 1980 (reference 3, Appendix A).	No photochemical skin reaction occurred under test condition. Ethanol solutions (25% w/v) of A13-36326-c caused a mild primary skin irritation to both the irradiated and nonirradiated skin areas.	Ethanol solutions of AI3-36326 are not expected to cause a photochemical irritant reaction in humans. However, ethanol solutions with batch AI3-36326-c may cause primary skin irritation.
ORAL LD_{50} STUDIES		
Oral Administration of Technical Grade and Corn Oil Solutions of AI3-36326-e		
Technical Grade AI3-36326-e		
Rats - male Six rats per dosage level. Treatment dosages ranged from 2000 to 7940 mg/Kg.	LD_{50}-5220 mg/Kg (95 % C.L. 4220-6470 mg/Kg) slope 5.89 SE $\pm$ 1.62. Signs were ataxia, bloody discharge from nose and mouth.	Compound relatively nonhazardous from ingestion.

Test	Results	Interpretation
Rats - female Six rats per dosage level. Treatment dosages ranged from 2000 to 6310 mg/Kg.	LD_{50}-3720 mg/Kg (95% C.L. 3230-4290 mg/Kg) slope 6.58 SE ± 1.48. Signs were ataxia, bloody discharge from nose and mouth.	Compound moderately toxic, may be harmful if ingested. Female rats may be more sensitive to lethal dosages than male rats.
Corn Oil Solution of AI3-36326-e		
Rats - female Six rats per dosage level. Treatment dosages ranged from 2000 to 6310 mg/Kg.	LD_{50}-3190 mg/Kg (95% C.L. 2710-3760 mg/Kg) slope 8.89 SE ± 2.46. Signs were ataxia and ruffled pelt.	Diluent has no apparent effect on lethal dosage responses.
INTRAPERITONEAL (IP) LD_{50} STUDIES		
IP Administration of Corn Oil Solutions of AI3-36326-e		
Technical Grade AI3-36326-e		
Rats - male Eight rats per dosage level. Treatment dosages ranged from 300 to 1000 mg/Kg.	LD_{50} -640 mg/Kg (95% C.L. 570-710 mg/Kg slope 15.69 SE ± 4.67. Signs at lethal dosages were tremors and ataxia.	Compound moderately toxic by this route of administration.
Rats - female Six rats per dosage level. Treatment dosages ranged from 398 to 1000 mg/Kg.	LD_{50}-520 mg/Kg (95% C.L. 460-600 mg/Kg) slope 14.62 SE ± 5.52.	Compound moderately toxic by IP route. No apparent sex difference in response to lethal dosages by this route of administration.
DERMAL LD_{50} STUDIES		
Dermal Application of Technical Grade AI3-36326-e to Rabbits		
Groups of five male and five female rabbits with abraded skin were exposed to a single topically applied occluded dosage of 2000 mg/Kg for 24 hours.	No deaths occurred in either group. (LD_{50} >2000 mg/Kg) Moderate skin irritation on all rabbits progressing to scabs at 14 days. No changes at 14-day gross necropsy.	No further dermal LD_{50} studies necessary according to proposed guidelines when no deaths occur at 2 g/Kg dosage (reference 4, Appendix A).

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Test	Results	Interpretation
SENSITIZATION STUDIES		
Test conducted according to Toxicology SOP, Guinea Pig Skin Sensitization test, August 1981 (ref 3, Appendix A). Suspension (w/v) of test compounds were prepared for injection in a mixture containing 1 volume of propylene glycol and 29 volumes of saline.	Guinea pigs receiving and challenged with a 0.1% suspension (w/v) of A13-36326-c did not show any sensitization reactions. Positive control (DNCB) produced a strong sensitization reaction in 10/10 guinea pigs.	Test compound AI3-36326-e did not exhibit a strong sensitization potential in guinea pigs and is not expected to cause a sensitization reaction in humans.
MUTAGENICITY PLATE ASSAY**		
AI3-36326-c was examined for genetic activity in a series of *in vitro* microbial assays employing *Salmonella* indicator organisms. The compound was tested directly and in the presence of liver microsomal enzyme preparations from Aroclor-1254®-induced rats.	The test material was toxic to the strains TA-98 and TA-100 at 25 µl and 50 µl doses. It was also slightly toxic at 5 and 10 µl doses for the strains TA-1535 and TA-1537 and at 10 µl doses for TA-1538 and TA-100.	AI3-36326 is not considered mutagenic under these test conditions.
The dose range employed for the evaluation of this compound was from 0.005 µl to 50 µl per plate.	The results of the tests in the presence and absence of a rat liver activation system were negative.	
Five strains of *Salmonella typhimurium*, TA-1535, TA-1537, TA-1538, TA-98 and TA-100 were used in evaluating mutagenic potential.		

® Aroclor is a registered trademark of Monsanto Chemical Co., 800 N. Lindberg Boulevard, St Louis, Missouri. Use of trademark names does not imply endorsement by the US Army, but is intended only to assist in identification of a specific product.

** Work performed under contract by Litton Bionetics, Kensington, Maryland. LBI Project No. 20988; Genetics Assay Number 5236.

Test	Results	Interpretation
Enzyme Induction Studies		
Test conducted according to Toxicology SOP, Enzyme Induction Studies in Rats, October 1981 (reference 3, Appendix A).	The mean hexobarbitol (100 mg/Kg) sleeping times with standard duration for each treatment group was as follows: AI3-36326-e 54.8 ± 10.4 min (15 mg/Kg) AI3-36326-e 44.3 ± 8.9 min (50 mg/Kg)† AI3-36326-e 35.6 ± 7.1 min (100 mg/Kg)† Phenobarbitol 29.0 ± 11.8 min (100 mg/Kg)† Solvent Control 65.3 ± 8.1 min (1 mL/Kg corn oil) Sham Control 64.2 ± 6.7 min (needle stick)	The decrease in hexobarbitol sleeping time indicates AI3-36326 is a potential inducer of hepatic microsomal enzymes with an aparent dose response relationship. Additional studies should be conducted to delineate specific enzyme systems involved.

† Statistically significant in the test at the 0.05 level compared to those of the sham control.

8. DISCUSSION. The test compound N,N-dipropylcyclohexanecarboxamide, AI3-36326, has shown great entomological potential as an insect repellent and has been proposed for this use in various preparations. Moderate irritation reactions were seen on rabbit skin following application of the technical grade material and a 25 percent (w/v) ethanol solution (reference 5, Appendix A). Although rabbit skin will usually give a maximum response to applied chemicals, additional studies with various AI3-36326 preparations and/or formulations must be investigated in order to define the skin irritant parameters and potential of this compound. Corneal irritant responses varied, seemingly dependent upon batch sample. Nevertheless, the potential for eye irritation may exist for sensitive individuals. Immediate rinsing of the eyes with copious amounts of water should be effective in decreasing or eliminating occular irritation from this compound. Evaluation of the initial unwashed eye response showed that the damage is not permanent but reversible within 3-7 days. Data indicate no great risk exists from acute ingestion or dermal exposure to this compound. However, the action of this compound on the liver by the induction of microsomal enzymes is cause for concern. Additional studies should be performed to elicit information on the enzymatic parameters of this compound which will aid in determining possible risks from the use of AI3-36326 as an insect repellent.

9. CONCLUSIONS. It is concluded that N,N-dipropylcyclohexanecarboxamide does not present an acute toxic hazard from accidental exposure. Various batch formulations present different eye and skin irritant responses.

10. RECOMMENDATIONS. The following recommendations are based upon professional judgement of the investigators.

a. Recommend consideration for further evaluation of this compound to include the following areas of emphasis.

(1) Continuation of subchronic studies to evaluate 21-day rabbit dermal responses and rodent teratology responses.

(2) Development of a test protocol to evaluate potential microsomal enzyme induction mechanisms.

(3) Development of a standard production formulation and characterization of specific chemical content.

(4) Development of a biological assay to evaluate skin irritation responses between different production lots.

b. Recommend immediate eye rinse with copious amounts of water to prevent or diminish ocular irritation in the unlikely event of unplanned human exposure.

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Biologist
Toxicology Division

MAURICE H. WEEKS
Chief, Toxicology Evaluation Branch
Toxicology Division

APPROVED:

TIMOTHY B. WEYANDT, M.D., M.P.H.
MAJ, MC
Acting Chief, Toxicology Division

APPENDIX A

REFERENCES

1. Letter, Agriculture Research, Northeastern Region, 14 February 1980, subject: Requested Synthesis Procedure.

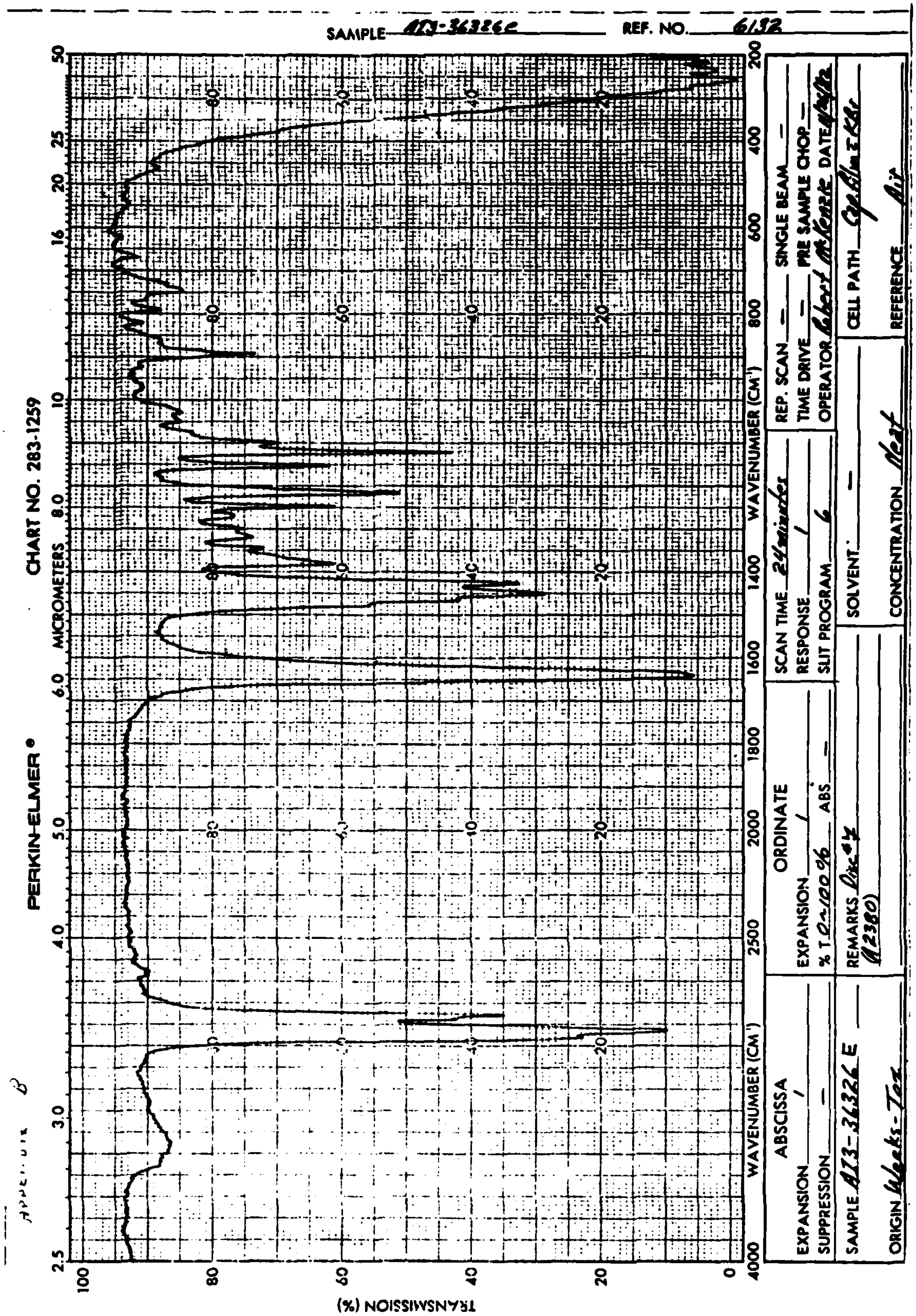
2. Toxicology Division Procedural Guide, US Army Environmental Hygiene Agency (USAEHA), 1972, revised 1976.

3. Toxicology Division Standing Operating Procedures, US Army Environmental Hygiene Agency (USAEHA) 1980-1982.

4. Proposed Guidelines for Registering Pesticides in the United States; Hazard Evaluation: Human and Domestic Animals, 43 Federal Register (FR) 37336, 22 August 1978.

5. Letter, HSE-LT/WP, this Agency, 18 July 1980, subject: Topical Hazard Evaluation Program of Candidate Insect Repellents AI3-36325, 1-(cyclohexylcarbonyl) hexahydro-1H-Azepine, A13-36326, N,N-dipropylcyclohexanecarboxamide, and A13-36328, 1-[(6-methyl-3-cyclohexen-1-yl) carbonyl] - Pyrrolidine, Study Nos. 75-51-0833-80, 75-51-0834-80, 75-51-0835-80, October 1975 - April 1980.

APPENDIX B


SAMPLE ATS-36326E
REF. NO. 6132
PERKIN-ELMER
CHART NO. 283-1259
MICROMETERS
WAVENUMBER (CM⁻¹)
TRANSMISSION (%)
ABSCISSA
EXPANSION 1
SUPPRESSION —
ORDINATE
EXPANSION 1
% T 0-100%
ABS —
SCAN TIME 24 minutes
RESPONSE 1
SLIT PROGRAM 6
REP. SCAN —
TIME DRIVE —
OPERATOR
SINGLE BEAM —
PRE SAMPLE CHOP —
DATE
SAMPLE ATS-36326 E
ORIGIN
REMARKS
SOLVENT —
CONCENTRATION Neat
CELL PATH
REFERENCE Air

APPENDIX C

EVALUATION OF SKIN REACTIONS*

Erythema and Eschar Formation	
No erythema	0
Very slight erythema (barely perceptible)	1
Well defined erythema	2
Moderate-to-severe erythema	3
Severe erythema (beet redness to slight eschar formation)	4
Edema Formation	
No edema	0
Very slight (barely perceptible)	1
Slight edema (edges or area well defined by definite raising)	2
Moderate edema (edges raised approximately 1 mm)	3
Severe edema (raised more than 1 mm and extending beyond area of exposure)	4

* An individual irritation score is equal to the sum of the scores for edema formation and erythema and eschar formation.

APPENDIX D

SCALE FOR SCORING OCULAR LESIONS

---

1. Cornea

   a. Opacity-degree of density (most dense area taken for reading)
      - No opacity.....0
      - Scattered or diffuse area, details of iris clearly visible.....1
      - Easily discernible translucent areas, details of iris slightly obscured.....2
      - Opalescent areas, no details of iris visible, size of pupil barely discernible.....3
      - Opaque, iris invisible.....4

   b. Area of cornea involved
      - One quarter (or less) but not zero.....1
      - Greater than one quarter but less than one half.....2
      - Greater than one half but less than three quarters.....3
      - Greater than three quarters up to whole area.....4

   Score = (a) x (b) x (5) = Total max score = 80

2. Iris

   Values
   - Normal.....0
   - Folds above normal, congestion, swelling, circumcorneal injection (any or all of these or combination of any thereof) iris still reacting to light (sluggish reaction is positive).....1
   - No reaction to light, hemorrhage, gross destruction (any or all of these).....2

   Score = (a) x 5 Total max score = 10

3. Conjunctivae

   a. Redness (refers to palpebral and bulbar conjunctivae excluding cornea and iris)
      - Vessels normal.....0
      - Vessels definitely injected above normal.....1
      - More diffuse, deeper crimson red, individual vessels not easily discernible.....2
      - Diffuse beefy red.....3

b. Chemosis

No swelling.....0
Any swelling above normal (included nictitating membrane).....1
Obvious swelling with partial eversion of lids.....2
Swelling with lids about half closed.....3
Swelling with lids about half closed to completely closed.....4

c. Discharge

No discharge.....0
Any amount different from normal (does not include small amounts observed with moistening of the lids and hairs just adjacent to lids.....2
Discharge with moistening of the lids and hairs, and considerable area around the eye.....3

Score (a + b + c) x 2 Total max score = 20

The individual numerical scores for each eye to which a given compound has been applied are added together and then divided by the number of eyes used to obtain the score.

APPENDIX E

TOPICAL HAZARD EVALUATION PROGRAM
DEFINITIONS OF CATEGORIES OF COMPOUNDS BEING CONSIDERED FOR ACUTE SKIN APPLICATION

CATEGORY I - Compounds producing no primary irritation of the intact skin or no greater than mild primary irritation of the skin surrounding an abrasion.

CATEGORY II - Compounds producing mild primary irritation of the intact skin and the skin surrounding an abrasion.

CATEGORY III - Compounds producing moderate primary irritation of the intact skin and the skin surrounding an abrasion.

CATEGORY IV - Compounds producing moderate to severe primary irritation of the intact skin and of the skin surrounding an abrasion and, in addition, producing necrosis, vesiculation and/or eschars.

CATEGORY V - Compounds impossible to classify because of straining of the skin or other masking effects owing to physical properties of the compound.

EYE CATEGORIES:

A. Compounds noninjurious to the eye.

B. Compounds producing mild injury to the cornea.

C. Compounds producing mild injury to the cornea, and in addition some injury to the conjunctiva.

D. Compounds producing moderate injury to the cornea.

E. Compounds producing moderate injury to the cornea, and in addition producing some injury to the conjunctiva.

F. Compounds producing severe injury to the cornea and to the conjunctiva.

APPENDIX F

ANALYTICAL QUALITY ASSURANCE

The Analytical Quality Assurance Office certifies the following with regard to this study:

a. This study was conducted in accordance with:

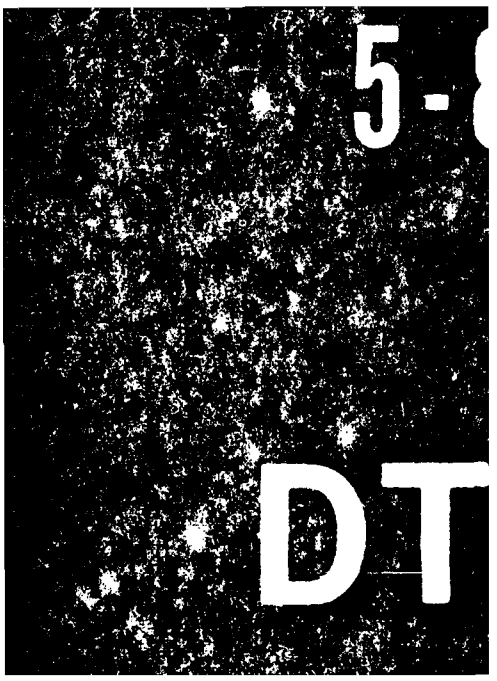
(1) Standing Operating Procedures developed by the Toxicology Division, USAEHA.

(2) Title 21, Code of Federal Regulations, 1981 rev, Part 58, Good Laboratory Practice for Nonclinical Laboratories Studies.

b. Facilities were inspected during its operational phase to insure compliance with paragraph a above.

c. The information presented in this report accurately reflects the raw data generated during the course of conducting the study.

PAUL V. SNEERINGER, Ph.D.
Chief, Analytical Quality
Assurance Office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