UNITED STATES ARMY
ENVIRONMENTAL HYGIENE
AGENCY

ABERDEEN PROVING GROUND, MD 21010

TOPICAL HAZARD EVALUATION PROGRAM
OF CANDIDATE INSECT REPELLENTS
US DEPARTMENT OF AGRICULTURE PROPRIETARY CHEMICALS
STUDY NUMBERS 75-51-0166-81, 75-51-0169-81 THRU 75-51-0171-81
AND 75-51-0177-81 THRU 75-51-0181-81
OCTOBER 1978 - JUNE 1981

Approved for public release; distribution unlimited.
### Abstract

Preliminary hazard evaluations of the above candidate insect repellent chemicals were performed by means of laboratory animal studies using rats, rabbits, and guinea pigs. Chemicals A13-37351a, 37352a, and 37564 were noninjurious to the eyes of rabbits. Chemical A13-37353a caused mild injury to the cornea of rabbits and chemicals A13-37347a, 37353a, 37560, 37561, and 37562 caused mild injury to the cornea and, in addition, some injury to the conjunctiva. All of the chemicals did not cause skin irritation with the exception of A13-37353a which caused mild skin irritation. All chemicals were relatively nontoxic by ingestion.
and did not cause photoirritation or prove to be skin sensitizers. Chemicals A13-37347a, 37352a, 37353a, 37563, and 37564 demonstrated some skin irritation from ethanol solutions during photoirritation studies. It was recommended that all chemicals be approved for further testing as candidate insect repellents.
DEPARTMENT OF THE ARMY
U. S. ARMY ENVIRONMENTAL HYGIENE AGENCY
ABERDEEN PROVING GROUND, MARYLAND 21010

SUBJECT: Topical Hazard Evaluation Program of Candidate Insect Repellents,
US Department of Agriculture Proprietary Chemicals, Study Numbers
75-51-0166-81, 75-51-0169-81 thru 75-51-0171-81, and 75-51-0177-81
thru 75-51-0181-81, October 1978 - June 1981

Executive Secretary
Armed Forces Pest Management Board
Forest Glen Section, WRAMC
Washington, DC 20012

A summary of the pertinent findings and recommendations of the inclosed
report follows:

Preliminary hazard evaluations of the above candidate insect repellent
chemicals were performed by means of laboratory animal studies using rats,
rabbits, and guinea pigs. Chemicals A13-37351a, 37352a, and 37564 were
noninjurious to the eyes of rabbits. Chemical A13-37563 caused mild injury
to the cornea of rabbits and chemicals A13-37347a, 37353a, 37560, 37561, and
37562 caused mild injury to the cornea and, in addition, some injury to the
conjunctiva. All of the chemicals did not cause skin irritation with the
exception of A13-37353a which caused mild skin irritation. All chemicals
were relatively nontoxic by ingestion and did not cause photoirritation or
prove to be skin sensitizers. Chemicals A13-37347a, 37352a, 37353a, 37563,
and 37564 demonstrated some skin irritation from ethanol solutions during
photoirritation studies. It was recommended that all chemicals be approved
for further testing as candidate insect repellents.

FOR THE COMMANDER:

[Signature]

JOHN F. MAZUR
LTC, MSC
Director, Laboratory Services

CF:
HQDA (DASG-PSP)
Cdr, HSC (HSPA-P)
Dir, Advisory Cen on Tox, NRC
Comdt, AHS (HSA-IPM)
USDA, ARS (Dr. Terrence McGovern)
USDA, ARS-Southern Region (2 cy)
TOPOCAL HAZARD EVALUATION PROGRAM
OF CANDIDATE INSECT REPELLENTS
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OCTOBER 1978 - JUNE 1981

1. AUTHORITY.

a. Letter, US Department of Agriculture - Agricultural Research Service,
   Southern Region, Insects Affecting Man Research Laboratory, Gainesville, FL,
   13 October 1978.

b. 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 Control Board; and the
   US Department of Agriculture, Agricultural Research, Science and Education
   Administration, titled, Coordination of Biological and Toxicological Testing

2. REFERENCE. Toxicology Division Standing Operating Procedures, US Army
   Environmental Hygiene Agency (USAEHA), 1981.

3. PURPOSE. The purpose of this program is to provide guidance for further
   entomological testing of the candidate insect repellents: A13-37347a,
   37351a, 37352a, 37353a, 37560, 37561, 37562, 37563, and 37564.

4. SUMMARY OF FINDINGS. Hazard evaluations of the above-named candidate
   repellents were conducted by this Agency using New Zealand White rabbits for
   skin and eye studies, Hartley guinea pigs for a skin sensitization study, and
   Sprague-Dawley rats for determination of oral toxicity. A tabular
   presentation of animal toxicity data developed in this Agency follows:* t

* 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) 78-23,
  revised 1978.
† The experiments reported herein were performed in animal facilities fully
  accredited by the American Association for the Accreditation of Laboratory
  Animal Care.

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<table>
<thead>
<tr>
<th>Test</th>
<th>Results</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>SKIN IRRITATION STUDIES</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Rabbits</strong></td>
<td></td>
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<tr>
<td>Single 24-hour application to intact and abraded skin of New Zealand White rabbits. 0.5 mL technical grade chemical applied to each of six rabbits.</td>
<td>Chemicals A13-37347a, 37351a, 37352a, 37560, 37561, 37562, 37563, and 37564 did not cause any irritation of the intact skin or of the skin surrounding an abrasion.</td>
<td>USAEHA Category I (ref Appendix A)</td>
</tr>
<tr>
<td></td>
<td>Chemical A13-37353a produced mild primary irritation of the intact skin and the skin surrounding an abrasion.</td>
<td>USAEHA Category II (ref Appendix A)</td>
</tr>
<tr>
<td><strong>EYE IRRITATION STUDIES</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Rabbits</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Single 24-hour application of 0.1 mL technical grade chemical to one eye of each of six New Zealand White rabbits.</td>
<td>Chemicals A13-37351a, 37352a, and 37564 did not cause any irritation to the eyes of rabbits.</td>
<td>USAEHA Category I (ref Appendix A)</td>
</tr>
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<td></td>
<td>Chemical A13-37563 caused mild injury to the cornea.</td>
<td>USAEHA Category B (ref Appendix A)</td>
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<tr>
<td></td>
<td>Chemicals A13-37347a, 3.353a, 37560, 37561, and 37562 caused mild injury to the cornea and, in addition, some injury to the conjunctiva.</td>
<td>USAEHA Category C (ref Appendix A)</td>
</tr>
</tbody>
</table>
Study Nos. 75-51-0166-81, 75-51-0169-81 thru 75-51-0171-81, and 75-51-0177-81 thru 75-51-0181-81, Oct 78 - Jun 81

Test Results Interpretation

APPROXIMATE LETHAL DOSE (ALD)

Oral

<table>
<thead>
<tr>
<th>Chemical</th>
<th>LD₅₀ (mg/Kg)</th>
<th>Interpretation</th>
</tr>
</thead>
<tbody>
<tr>
<td>A13-37347a</td>
<td>4300</td>
<td>These chemicals are relatively nontoxic by ingestion.</td>
</tr>
<tr>
<td>A13-37351a</td>
<td>4300</td>
<td></td>
</tr>
<tr>
<td>A13-37352a</td>
<td>9700</td>
<td></td>
</tr>
<tr>
<td>A13-37353a</td>
<td>9700</td>
<td></td>
</tr>
<tr>
<td>A13-37560</td>
<td>4300</td>
<td></td>
</tr>
<tr>
<td>A13-37561</td>
<td>9700</td>
<td></td>
</tr>
<tr>
<td>A13-37562</td>
<td>4300</td>
<td></td>
</tr>
<tr>
<td>A13-37563</td>
<td>6500</td>
<td></td>
</tr>
<tr>
<td>A13-37564</td>
<td>6500</td>
<td></td>
</tr>
</tbody>
</table>

PHOTOCHEMICAL SKIN IRRITATION STUDIES

Rabbits

A single 0.05 mL application of a 25-percent (w/v) solution of each chemical and a 10-percent (w/v) Oil of Bergamot solution (positive control) in 95-percent ethyl alcohol were applied to the intact skin of six rabbits. Five minutes after application, the rabbits were exposed to UV light (365 nm) for 30 minutes at a distance of 10-15 cm.

A 25-percent solution of each tested chemical in ethanol did not cause a photochemical irritation reaction under test conditions.

Ethanol solutions of A13-37347a, 37352a, 37353a, 37563, and 37564 caused irritation at both UV and non-UV skin sites.

All tested chemicals did not cause a photochemical irritation reaction under test conditions and are not expected to cause a photochemical irritation in humans. Ethanol solutions of A13-37347a, 37352a, 37353a, 37563, and 37564 may cause skin irritation in some sensitive individuals. Personnel experiencing this reaction should wash off the solution as soon as possible.

Control

Following UV exposures of the rabbits, 0.05 mL of test chemical, positive control and diluent were applied to additional skin areas to serve as unirradiated control sites. Application areas were checked for skin irritation at 24, 48 and 72 hours.

Positive control application and irradiation caused greater irritant effects than in unirradiated skin areas.
<table>
<thead>
<tr>
<th>Test</th>
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</tr>
</thead>
<tbody>
<tr>
<td><strong>SENSITIZATION STUDIES</strong>&lt;br&gt; <strong>Guinea Pigs (Male)</strong>&lt;br&gt;Intradermal injections of 0.1 mL of a 0.1-percent solution (w/v) of the tested chemicals or of dinitrochlorobenzene (DNCB)* in a mixture containing 1 volume of propylene glycol and 29 volumes of saline.&lt;br&gt;Ten test guinea pigs for each chemical were given 10 sensitizing doses over a 3-week period. After 2 weeks' rest, they were challenged with ID injections of each test chemical.&lt;br&gt;Ten positive control guinea pigs were sensitized over 3 weeks with DNCB. After 2 weeks' rest, they were challenged with ID injections of DNCB.</td>
<td>Challenge doses of the tested chemicals did not produce a sensitization reaction.&lt;br&gt;The tested chemicals did not produce sensitization reactions under test conditions and are not expected to produce sensitization reactions in man.&lt;br&gt;Challenge dose of DNCB in positive control guinea pigs produced a marked sensitization reaction in 10 out of 10 guinea pigs.</td>
<td>DNCB produced a marked reaction, indicating the guinea pigs respond to sensitizing agents.</td>
</tr>
</tbody>
</table>

* A known skin sensitizer.
Study Nos. 75-51-0166-81, 75-51-0169-81 thru 75-51-0171-81, and 75-51-0177-81 thru 75-51-0181-81, Oct 78 - Jun 81

5. CONCLUSION. Chemicals A13-37351a, 37352a, and 37564 were noninjurious to the eyes of rabbits. Chemical A13-37563 caused mild injury to the cornea of rabbits, and chemicals A13-37347a, 37353a, 37560, 37561, and 37562 caused mild injury to the cornea and, in addition, some injury to the conjunctiva. All of the chemicals did not cause skin irritation with the exception of A13-37353a which caused mild skin irritation. All chemicals were relatively nontoxic by ingestion and did not cause photoirritation or prove to be skin sensitizers. Chemicals A13-37347a, 37352a, 37353a, 37563, and 37564 demonstrated some skin irritation from ethanol solutions during photoirritation studies.

6. RECOMMENDATION. Under the provisions of the Memorandum of Understanding (paragraph 1b), it is recommended that the following USDA proprietary chemicals be approved for further testing as candidate insect repellents: A13-37347a, 37351a, 37352a, 37353a, 37560, 37561, 37562, 37563, and 37564. Ethanol solutions of chemicals A13-37347a, 37352a, 37353a, 37563, and 37564 may cause skin irritation in sensitive individuals and, if experienced, the site should be washed with copious amounts of water.

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CPT, VC
Laboratory Animal Veterinary Officer
Toxicology Division

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APPROVED:

ARTHUR H. MCCREESH, Ph.D.
Chief, Toxicology Division
APPENDIX A

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.
(INTERPRETATION: No restriction for acute application to the human skin.)

CATEGORY II - Compounds producing mild primary irritation of the intact skin
and the skin surrounding an abrasion. (INTERPRETATION: Should be used only
on human skin found by examination to have no abrasions or may be used as a
clotting impregnant.)

CATEGORY III - Compounds producing moderate primary irritation of the intact
skin and the skin surrounding an abrasion. (INTERPRETATION: Should not be
used directly on the skin without a prophetic patch test having been
conducted on humans to determine irritation potential to human skin. May be
used without patch testing, with extreme caution, as clothing impregnants.
Compound should be resubmitted in the form and at the intended use
concentration so that its irritation potential can be reexamined using other
test techniques on animals.)

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. (INTERPRETATION: Should
be resubmitted for testing in the form and at the intended use concentration.
Upon resubmission, its irritation potential will be reexamined using other
test techniques on animals, prior to possible prophetic patch testing in
humans, at concentrations which have been shown not to produce primary
irritation in animals.)

CATEGORY V - Compounds impossible to classify because of staining of the skin
or other masking effects owing to physical properties of the compound.
(INTERPRETATION: Not suitable for use on humans.)

EYE CATEGORIES:

A. Compounds noninjurious to the eye. INTERPRETATION: Irritation of
human eyes is not expected if the compound should accidentally get into the
eyes, provided it is washed out as soon as possible.

B. Compounds producing mild injury to the cornea. INTERPRETATION:
Should be used with caution around the eyes.

C. Compounds producing mild injury to the cornea, and in addition some
injury to the conjunctiva. INTERPRETATION: Should be used with caution
around the eyes and mucosa.

D. Compounds producing moderate injury to the cornea. INTERPRETATION:
Should be used with extreme caution around the eyes.

E. Compounds producing moderate injury to the cornea, and in addition
producing some injury to the conjunctiva. INTERPRETATION: Should be used
with extreme caution around the eyes and mucosa.

F. Compounds producing severe injury to the cornea and to the
conjunctiva. INTERPRETATION: Should be used with extreme caution. It is
recommended that use be restricted to areas other than the face.
Study Nos. 75-51-0166-81, 75-51-0169-81 thru 75-51-0171-81, and 75-51-0177-81 thru 75-51-0181-81, Oct 78 - Jun 81

APPENDIX B

ANALYTICAL QUALITY ASSURANCE

The Analytical Quality Assurance Office certifies the following with regard to the Topical Hazard Evaluation Program of Candidate Insect Repellents, US Department of Agriculture Proprietary Chemicals, Study Numbers 75-51-0166-81, 75-51-0169-81 thru 75-51-0171-81, and 75-51-0177-81 thru 75-51-0181-81, October 1978 - June 1981.

a. This study was conducted in accordance with:

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


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

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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