



ORIGINAL FILE COPY

6

Institute Report No. 413

AD-A215 145

# Primary Dermal Irritation Potential of JA-2 Solid Propellant in Rabbits

*Earl W. Morgan, DVM, MAJ, VC*  
*James D. Justus, MPA, SSG, USA*  
*and*  
*Don W. Korte, Jr., PhD, LTC, MSC*

MAMMALIAN TOXICOLOGY BRANCH  
DIVISION OF TOXICOLOGY

DTIC  
ELECTE  
DEC 12 1989  
S  
D

October 1989

Toxicology Series: 175

DISTRIBUTION STATEMENT A  
Approved for public release;  
Distribution Unlimited

LETTERMAN ARMY INSTITUTE OF RESEARCH  
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129

89 12 11 082

Primary Dermal Irritation Potential of JA-2 Solid Propellant in Rabbits (Toxicology Series 175)--Morgan *et al.*

This document has been approved for public release and sale; its distribution is unlimited.

Destroy this report when it is no longer needed. Do not return to the originator.

Citation of trade names in this report does not constitute an official endorsement or approval of the use of such items.

This research was conducted in compliance with the "Guide for the Care and Use of Laboratory Animals," NIH Publication No. 85-23, as prepared by the Institute of Laboratory Animal Resources, National Research Council.

This material has been reviewed by Letterman Army Institute of Research and there is no objection to its presentation and/or publication. The opinions or assertions contained herein are the private views of the author(s) and are not to be construed as official or as reflecting the views of the Department of the Army or the Department of Defense. (AR 360-5)

*Donald G. Corby* 30 Oct 89  
\_\_\_\_\_  
Donald G. Corby (date)  
COL, MC  
Commanding

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE

## REPORT DOCUMENTATION PAGE

Form Approved  
OMB No. 0704-0188

|  |  |  |  |
|--|--|--|--|
| 1a. REPORT SECURITY CLASSIFICATION<br>UNCLASSIFIED   |  | 1b. RESTRICTIVE MARKINGS   |  |
| 2a. SECURITY CLASSIFICATION AUTHORITY  |  | 3. DISTRIBUTION / AVAILABILITY OF REPORT   |  |
| 2b. DECLASSIFICATION / DOWNGRADING SCHEDULE  |  | APPROVED FOR PUBLIC RELEASE;<br>DISTRIBUTION IS UNLIMITED.                                       |  |
| 4. PERFORMING ORGANIZATION REPORT NUMBER(S)<br><br>Institute Report No.: 413   |  | 5. MONITORING ORGANIZATION REPORT NUMBER(S)  |  |
| 6a. NAME OF PERFORMING ORGANIZATION<br>Mammalian Toxicology<br>Division of Toxicology  | 6b. OFFICE SYMBOL<br>(If applicable)<br>SGRD-ULE-T | 7a. NAME OF MONITORING ORGANIZATION<br>US Army Biomedical Research<br>and Development Laboratory |  |
| 6c. ADDRESS (City, State, and ZIP Code)<br>Letterman Army Institute of Research<br>Presidio of San Francisco, CA 94129-6800  |  | 7b. ADDRESS (City, State, and ZIP Code)<br>Fort Detrick<br>Frederick, MD 21701-5010              |  |
| 8a. NAME OF FUNDING / SPONSORING<br>ORGANIZATION US Army Medical<br>Research & Development Command   | 8b. OFFICE SYMBOL<br>(If applicable)               | 9. PROCUREMENT INSTRUMENT IDENTIFICATION NUMBER  |  |
| 8c. ADDRESS (City, State, and ZIP Code)<br>Fort Detrick<br>Frederick, Maryland 21701-5012  |  | 10. SOURCE OF FUNDING NUMBERS  |  |
|  |  | PROGRAM<br>ELEMENT NO.<br>62720  | PROJECT<br>NO.<br>A835   |
|  |  | TASK<br>NO.<br>AB  | WORK UNIT<br>ACCESSION NO.<br>DA303913   |
| 11. TITLE (Include Security Classification)<br>(U) Primary Dermal Irritation Potential of JA-2 Solid Propellant in Rabbits   |  |  |  |
| 12. PERSONAL AUTHOR(S)<br><br>EW Morgan, JD Justus, and DW Korte, Jr.  |  |  |  |
| 13a. TYPE OF REPORT<br>Institute   | 13b. TIME COVERED<br>FROM 14 NOV 85 to 7 DEC 85    | 14. DATE OF REPORT (Year, Month, Day)<br>October 1989  | 15. PAGE COUNT<br>17   |
| 16. SUPPLEMENTARY NOTATION<br><br>Toxicology Series No. 175  |  |  |  |
| 17. COSATI CODES   |  | 18. SUBJECT TERMS (Continue on reverse if necessary and identify by block number)                |  |
| FIELD  | GROUP  | SUB-GROUP  |  |
|  |  |  | 7 JA-2 Solid Propellant; Primary Dermal Irritation;<br>Mammalian Toxicology; Nitroglycerin; Munitions;<br>Diethyleneglycol Dinitrate; Rabbits (U) 6- |
| 19. ABSTRACT (Continue on reverse if necessary and identify by block number)<br>The primary dermal irritation potential of JA-2 Solid Propellant was determined in female New Zealand White rabbits by using a modified Draize method. Very slight erythema and edema were observed in 1 of 8 rabbits one hour after dosing. This rabbit had returned to normal by 24 hours after dosing. No other recognizable skin reaction was detected at any time during the 14-day observation period. The test compound was non-irritating under conditions of this study. <i>Key words: toxicity</i> |  |  |  |
| 20. DISTRIBUTION / AVAILABILITY OF ABSTRACT<br><input checked="" type="checkbox"/> UNCLASSIFIED/UNLIMITED <input type="checkbox"/> SAME AS RPT. <input type="checkbox"/> DTIC USERS  |  | 21. ABSTRACT SECURITY CLASSIFICATION<br>UNCLASSIFIED   |  |
| 22a. NAME OF RESPONSIBLE INDIVIDUAL<br>DONALD G. CORBY, COL, MC  |  | 22b. TELEPHONE (Include Area Code)<br>(415) 561-3600   | 22c. OFFICE SYMBOL<br>SGRD-ULZ   |

### ABSTRACT

The primary dermal irritation potential of JA-2 Solid Propellant was determined in female New Zealand White rabbits by using a modified Draize method. Very slight erythema and edema were observed in 1 of 8 rabbits one hour after dosing. This rabbit had returned to normal by 24 hours after dosing. No other recognizable skin reaction was detected at any time during the 14-day observation period. The test compound was non-irritating under conditions of this study.

**KEY WORDS:** Primary Dermal Irritation, JA-2 Solid Propellant, Mammalian Toxicology, Rabbit, Munition, Nitroglycerin, Diethyleneglycol dinitrate

|                      |                                     |
|----------------------|-------------------------------------|
| <b>Accession For</b> |                                     |
| DTIS GRA&I           | <input checked="" type="checkbox"/> |
| DTIC TAB             | <input type="checkbox"/>            |
| Unannounced          | <input type="checkbox"/>            |
| Justification        |                                     |
| By _____             |                                     |
| Distribution/        |                                     |
| Availability Codes   |                                     |
| Dist                 | Avail and/or<br>Special             |
| A-1                  |                                     |



## PREFACE

**TYPE REPORT:** Primary Dermal Irritation GLP Study Report

**TESTING FACILITY:**

US Army Medical Research and Development Command  
Letterman Army Institute of Research  
Presidio of San Francisco, CA 94129-6800

**SPONSOR:**

US Army Medical Research and Development Command  
US Army Biomedical Research and Development Laboratory  
Fort Detrick, Maryland 21701-5010  
Project Officer: Gunda Reddy, PhD

**PROJECT/WORK UNIT/APC:** 3E162720A835/180/TLB0

**GLP STUDY NUMBER:** 85019

**STUDY DIRECTOR:** LTC Don W. Korte, Jr., PhD, MSC  
Diplomate, American Board of Toxicology

**PRINCIPAL INVESTIGATOR:** MAJ Earl W. Morgan, DVM, VC, Diplomate,  
American College of Veterinary Preventive  
Medicine, American Board of Toxicology

**CO-INVESTIGATOR:** SSG James D. Justus, MPA, USA

**PATHOLOGIST:** MAJ G. Tracey Makovec, DVM, VC, Diplomate,  
American College of Veterinary Pathologists

**REPORT AND DATA MANAGEMENT:**

A copy of the final report, study protocol, retired SOPs, raw data, analytical, stability, and purity data of the test compound, and an aliquot of the test compound will be retained in the LAIR Archives.

**TEST SUBSTANCE:** JA-2 Solid Propellant

**INCLUSIVE STUDY DATES:** 14 November - 17 December 1985

**OBJECTIVE:** The objective of this study was to determine the primary dermal irritation potential of JA-2 Solid Propellant in female New Zealand White rabbits.

## **ACKNOWLEDGMENTS**

SP4 James J. Fischer, SP4 Scott L. Schwebe, and SP4 Theresa L. Polk provided technical assistance; SP4 Paul B. Simboli provided assistance for the chemical analysis; Diane Arevelo and Obie Goodrich provided care for the animals; and Colleen S. Kamiyama provided secretarial assistance.

**SIGNATURES OF PRINCIPAL SCIENTISTS INVOLVED IN THE STUDY**

We, the undersigned, declare that GLP Study 85019 was performed under our supervision, according to the procedures described herein, and that this report is an accurate record of the results obtained.

*Don W. Korte Jr 30 Oct 89*

DON W. KORTE JR., PhD / DATE  
LTC, MSC  
Study Director

*Earl W. Morgan 2 Jul 89*

EARL W. MORGAN, DVM / DATE  
MAJ, VC  
Principal Investigator

*James D. Justus 2 Aug 89*

JAMES D. JUSTUS, MPA / DATE  
SSG, USA  
Co-Principal Investigator

*Conrad R. Wheeler 30 Oct 89*

CONRAD R. WHEELER, PhD / DATE  
DAC  
Analytical Chemist



DEPARTMENT OF THE ARMY  
LETTERMAN ARMY INSTITUTE OF RESEARCH  
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129-6000

REPLY TO  
ATTENTION OF:

SGRD-ULZ-QA

26 October 1989

MEMORANDUM FOR RECORD

SUBJECT: GLP Compliance for GLP Study 85019

1. This is to certify that the protocol for LAIR GLP Study 85019 was reviewed on 10 May 1985.
2. The institute report entitled "Primary Dermal Irritation Potential of JA-2 Solid Propellant in Rabbits," Toxicology Series 175, was audited on 25 October 1989.

*Carolyn M. Lewis*

CAROLYN M. LEWIS  
Diplomate, American Board of  
Toxicology  
Quality Assurance Auditor

## TABLE OF CONTENTS

|   |     |
|---|-----|
| Abstract .....                                      | i   |
| Preface .....                                       | iii |
| Acknowledgments.....                                | iv  |
| Signatures of Principal Scientists.....             | v   |
| Report of Quality Assurance Unit.....               | vi  |
| Table of Contents.....                              | vii |
| <br>  |     |
| INTRODUCTION.....                                   | 1   |
| Objective of Study.....                             | 1   |
| <br>  |     |
| MATERIALS.....                                      | 1   |
| Test Substance.....                                 | 1   |
| Animal Data.....                                    | 2   |
| Husbandry.....                                      | 2   |
| <br>  |     |
| METHODS.....  | 2   |
| Group Assignment/Acclimation.....                   | 2   |
| Test Procedures.....                                | 3   |
| Observations.....                                   | 3   |
| Duration of Study.....                              | 3   |
| Changes/Deviations.....                             | 3   |
| Storage of Raw Data and Final Report.....           | 5   |
| <br>  |     |
| RESULTS.....  | 5   |
| <br>  |     |
| DISCUSSION.....                                     | 5   |
| <br>  |     |
| CONCLUSION.....                                     | 6   |
| <br>  |     |
| REFERENCES.....                                     | 7   |
| <br>  |     |
| APPENDICES.....                                     | 8   |
| Appendix A. Chemical Data.....                      | 9   |
| Appendix B. Animal Data.....                        | 12  |
| Appendix C. Historical Listing of Study Events..... | 13  |
| Appendix D. Dermal Irritation Data.....             | 14  |
| Appendix E. Pathology Report.....                   | 16  |
| <br>  |     |
| OFFICIAL DISTRIBUTION LIST.....                     | 17  |

**Primary Dermal Irritation Potential of JA-2 Solid Propellant in Rabbits—  
Morgan et al.**

**INTRODUCTION**

The Department of Defense is considering the use of diethyleneglycol dinitrate (DEGDN), triethyleneglycol dinitrate (TEGDN), or trimethylolethane trinitrate (TMETN) as a replacement for nitroglycerin in new propellant formulations. However, considerable gaps in the toxicology data of the compounds were identified during a review of their health effects (1) conducted for the US Army Biomedical Research and Development Laboratory (USABRDL). Consequently, USABRDL has tasked the Division of Toxicology, Letterman Army Institute of Research (LAIR), to conduct an initial health effects evaluation of the proposed replacement nitrate esters. This initial evaluation of DEGDN, TMETN, TEGDN, and two DEGDN-based propellants, JA-2 and DIGL-RP, includes the Ames mutagenicity assay, acute oral toxicity tests in rats and mice, acute dermal toxicity in rabbits, dermal and ocular irritation studies in rabbits, and dermal sensitization studies in guinea pigs.

Objective of Study

The objective of this study was to determine the primary dermal irritation potential of JA-2 Solid Propellant in female New Zealand White rabbits.

**MATERIALS**

Test Substance

Chemical Name: JA-2 Solid Propellant

LAIR Code Number: TP56

Morgan et al.-2

Description: Solid black cylinders (stick configuration)

Lot Number: RAD83K001S153

JA-2 Solid Propellant was received in the stick configuration. It was ground into a fine powder for the study (Appendix A). Other test substance information is presented in Appendix A.

#### Animal Data

Eight female New Zealand White rabbits (Elkhorn Rabbitry, Watsonville, CA), identified individually with ear tattoos numbered 85F301 to 85F308 inclusive, were assigned to the study. The animal weights on dosing day (3 Dec 85) ranged from 2.5 to 3.0 kg. Additional animal data appear in Appendix B.

#### Husbandry

The rabbits were housed individually in stainless steel, screen-bottomed, battery-type cages with automatically flushing dump tanks. The diet consisted of 150 g per day of Certified Purina Chow<sup>®</sup> Diet 5322 (Ralston Purina Company, Checkerboard Square, St. Louis, MO); water was provided by continuous drip from a central line. The animal room temperature was maintained at 17.0° to 19.5°C with a relative humidity range of 50 to 65 percent with short spikes up to 78 percent associated with room cleaning. The photoperiod was 12 hours of light per day.

### **METHODS**

#### Group Assignment/Acclimation

Study animals were acclimated for 6 days to the study room following a 14-day quarantine by the Division of Animal Care and Services. During this period they were observed daily for signs of illness. They were treated prophylactically for ear mites with a single dose of Canex<sup>®</sup> and mineral oil instilled in the ears.

### Test Procedures

This study was conducted in accordance with EPA guidelines (2) and LAIR SOP-OP-STX-34 (3).

The backs of 8 rabbits were close-clipped 24 hours before the actual dosing. The clipped area was divided into 4 quadrants designated HV (4, 5). Site I was a sham patch control site. Sites II and III were test compound sites. Site IV was treated with an isotonic saline control patch. A standard dose of 0.5 g of powdered JA-2 was mixed with 0.5 ml isotonic saline (Viaflex<sup>®</sup>, Sodium Chloride Injection, USP; Travenol Laboratories, Inc., Deerfield, IL) to make a paste which was then placed on 1-inch (2.5 cm) square gauze patches that were taped to the appropriate sites. Blenderm<sup>®</sup> (Medical Products Division of 3M, Saint Paul, MN), a semi-impervious, hypoallergenic surgical tape, was used to hold the patches in place. Vet Wrap<sup>®</sup> (Animal Care Products Division of 3M, Saint Paul, MN) was then wrapped securely around the animal. The test compound was left in contact with the skin for 4 hours. At the end of the exposure period the wrapping and patches were removed, and the areas were scored one hour later.

### Observations

The grading and scoring for dermal reactions were performed according to Table 1. Scoring and grading were performed at approximately 1, 24, 48, and 72 hours, and 7 and 14 days after removal of the patch. Observations for clinical signs were made daily from 3 to 17 December 1985. After 14 days the animals were submitted for necropsy.

### Duration of Study

Appendix C is a complete historical listing of study events.

### Changes/Deviations

Ventilation fans in the building were turned off for several hours on 7 Dec 85. This resulted in a temperature and humidity increase in the animal room. The temperature rose to 20°C and the humidity rose to 79% for a

---

**TABLE 1 (4)**  
**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 [injurious in depth]) | 4        |
| <b>Possible total erythema score</b>   | <b>4</b> |

**Edema Formation**

|  |          |
|--|----------|
| No edema   | 0        |
| Very slight edema (barely perceptible)                                     | 1        |
| Slight edema (edges of 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        |
| <b>Possible total edema score</b>  | <b>4</b> |

**Possible total score for primary irritation** **8**

---

period of approximately four hours. This is not believed to have had any effect on the study.

#### Storage of Raw Data and Final Report

A copy of the final report, study protocols, raw data, retired SOPs, and an aliquot of the test compound were retained in the LAIR Archives.

#### **RESULTS**

Animals were scored for erythema and edema at each patch site. Rabbit 85F307 was observed to have very slight erythema and edema one hour after dosing at both test compound sites. The skin at the sham patch site on animal 85F301 had slight erythema at one hour after patch removal. Both animals returned to normal by 24 hours after dosing and remained normal throughout the study. No other recognizable skin reaction was detected at any time during the 14-day observation period. The vehicle control patch sites were normal throughout the study. Total scores (erythema plus edema) for the dermal irritation potential in each rabbit were tabulated (Appendix D). Fourteen days after topical application there were no gross lesions that could be attributed to exposure to the test material (Appendix E).

#### **DISCUSSION**

The modified Draize dermal irritation test as performed for this study has proven reliable for detecting non-irritating substances and severe irritants but considerably less reliable for detecting mild and moderate irritants (5). Consequently, many systems have been used to score and categorize the dermal irritation potential of a test compound. The system used by the Toxicity Testing Program at LAIR is an adaptation of one used at the U.S. Army Environmental Hygiene Agency (6). It develops a dermal irritation index based on the peak net mean score, which is the maximum net mean score calculated during the 72-hour observation period. Non-irritating compounds have peak net mean scores of 0.0 to 0.5. Mild irritants have peak net mean

Morgan et al.-6

scores of 0.51 to 2.0. Moderate irritants have peak net mean scores of 2.1 to 5.0. Severe irritants have peak net mean scores of 5.1 to 8.0. JA-2 Solid Propellant produced very slight erythema and edema in 1 of 8 rabbits. The peak net mean score for the test compound was 0.0; therefore, JA-2 was classified as a non-irritant.

#### **CONCLUSION**

The test compound, JA-2 Solid Propellant, is not a dermal irritant under conditions of this assay.

## REFERENCES

1. Holleman JW, Ross RH, Carroll JW. Problem definition study on the health effects of diethyleneglycol dinitrate, triethyleneglycol dinitrate, and trimethylolethane trinitrate and their respective combustion products. Frederick, MD: US Army Medical Bioengineering Research and Development Laboratory, 1983, DTIC No. ADA 127846.
2. Environmental Protection Agency. Office of Pesticide and Toxic Substances, Office of Toxic Substances (TS-792). Primary dermal irritation. In: Health effects test guidelines. Washington, DC: Environmental Protection Agency, August 1982; EPA 560/6-82-001.
3. Primary dermal irritation study. LAIR Standard Operating Procedure OP-STX-34, Presidio of San Francisco, CA: Letterman Army Institute of Research, 1 August 1984.
4. Draize JH, Woodard G, Calvery HO. Methods for the study of irritation and toxicity of substances applied topically to the skin and mucous membranes. *J Pharmacol Exp Ther* 1944; 83:377-390.
5. McCreesh AH, Steinberg M. Skin irritation testing in animals. In: Marzulli FN, Maibach HI, eds. *Dermatotoxicology*. 3rd ed. Washington, DC: Hemisphere Publishing Corp, 1987: 153-172.
6. US Army Environmental Hygiene Agency. Topical hazard evaluation program. Procedural guide. Aberdeen Proving Ground, MD: US Army Environmental Hygiene Agency, October 1985.

|   |    |
|---|----|
| Appendix A. Chemical Data.....                      | 9  |
| Appendix B. Animal Data.....                        | 12 |
| Appendix C. Historical Listing of Study Events..... | 13 |
| Appendix D. Dermal Irritation Data .....            | 14 |
| Appendix E. Pathology Report.....                   | 16 |

### Appendix A: CHEMICAL DATA

Test substance: JA-2 Solid Propellant

LAIR Code Number: TP56

Physical State: Solid black cylinders (stick configuration)

Preparation of test substance for dosing: The cylinders of JA-2 were ground to a fine powder under liquid nitrogen using a Spex freezer mill. The powder was then sieved through an 80-mesh screen.

#### Chemical Analysis:

DEGDN was the only major component of JA-2 which could be easily analyzed.<sup>1</sup> To determine the percent DEGDN in the JA-2 propellant, samples of JA-2 powder were added to individual 100 ml volumetric flasks. After dilution to volume with 95% ethanol, a second 1:100 dilution was performed. These solutions were analyzed by HPLC. Standards consisted of solutions of DEGDN in ethanol ranging in concentration from 164.5 to 670.5 µg/ml. Analysis of DEGDN by HPLC was performed under the following conditions: column, Brownlee RP-18 (4.6 x 250 mm, Brownlee Labs, Inc., Santa Clara, CA); solvent system, 40% water - 60% acetonitrile; flow rate, 0.9 ml/min; wavelength monitored, 210 nm.<sup>2</sup> Under these conditions, DEGDN eluted with a retention time of approximately 5.4 min.

The results from the analysis of standards and JA-2 powder samples are presented in Tables 1 and 2.

Table 1. Analysis of standards

| Concentration of Standard (µg/ml) | Peak Area* (x 10 <sup>-7</sup> ) |
|-----------------------------------|----------------------------------|
| 164.5                             | 0.94                             |
| 191.0                             | 1.09                             |
| 275.5                             | 1.60                             |
| 299.4                             | 1.74                             |
| 362.0                             | 2.08                             |
| 399.6                             | 2.31                             |
| 444.4                             | 2.52                             |
| 539.8                             | 3.07                             |
| 585.0                             | 3.32                             |
| 670.5                             | 3.79                             |

\*Average of 2 determinations

Equation for line by linear regression analysis:

$$Y = 5.62 \times 10^{-4} X + 3.51 \times 10^{-5}, r^2 = 0.9999$$

## Appendix A (cont.): CHEMICAL DATA

Table 2. Analysis of JA-2 Powder

| Weight of JA-2 Analyzed (mg) | Dilution Factor | Peak Area (x 10 <sup>-7</sup> ) | Conc of DEGDN in JA-2 (weight %)* |
|------------------------------|-----------------|---------------------------------|-----------------------------------|
| 104.8                        | 100             | 1.56                            | 25.9                              |
| 101.6                        | 100             | 1.57                            | 26.9                              |
| 109.7                        | 100             | 1.69                            | 26.8                              |

\*Calculated using the equation for the standard curve as follows:  

$$= \{[(\text{Peak Area} - 3.51 \times 10^5) / 5.62 \times 10^4] + \text{wgt JA-2 (mg)}\} \times 10.$$

The average value for the concentration of DEGDN in JA-2 was 27% and this agrees closely with the value of  $24.82 \pm 1.50$  % reported in the data sheet provided by the source.

**Stability:** The aqueous stability of the DEGDN component of JA-2 propellant was determined.<sup>3</sup> The amount of DEGDN in JA-2 suspensions was determined immediately after preparation of a suspension and again 24 hours later. The study was conducted as follows: A suspension of JA-2 in 1% gum tragacanth (200 mg/ml) was prepared. Three 1 ml aliquots were removed from the suspension immediately after preparation and again 24 hours later. The 1 ml samples were transferred to individual 100 ml volumetric flasks. After diluting to volume with ethanol, the solutions were analyzed by HPLC as described above. The average of the peak area values was  $2.92 \pm 0.12$  for the 0 time samples and  $2.95 \pm 0.11$  for the 24-hour samples. These results indicate that there was no decomposition of DEGDN in 1% gum tragacanth for a period of 24 hours.

**Source:** Radford Army Ammunition Plant, Radford, Virginia  
 (prime contractor: Hercules Inc, Wilmington, Delaware)

Lot no.: RAD83K001S153

<sup>1</sup> Wheeler CR. Toxicity Testing of Propellants. Laboratory Notebook #85-12-023, p. 51-61. Letterman Army Institute of Research, Presidio of San Francisco, CA.

<sup>2</sup> Wheeler CW. Nitrocellulose-Nitroguanidine Projects. Laboratory Notebook #84-05-010.3, p. 58. Letterman Army Institute of Research, Presidio of San Francisco, CA.

<sup>3</sup> Wheeler CR. Toxicity Testing of Propellants. Laboratory Notebook #85-12-023, p. 27, 35, 41. Letterman Army Institute of Research, Presidio of San Francisco, CA.

**Appendix A (cont.): CHEMICAL DATA****CHEMICAL ANALYSIS FOR JA-2  
(Information from the Manufacturer's Data Sheet)**

| <u>Ingredient</u>  | <u>Finished Propellant<br/>Percentage</u> |
|--|---|
| Nitrocellulose<br>(13.8% $\pm$ 0.05% Nitrogen)<br>(6-12 seconds viscosity) | 58.5 $\pm$ 2.00                           |
| Nitroglycerin  | 15.88 $\pm$ 1.00                          |
| Diethyleneglycol dinitrate (DEGDN)   | 24.82 $\pm$ 1.50                          |
| Akardit II   | 0.70 $\pm$ 0.20                           |
| Magnesium Oxide  | 0.04 Max                                  |
| Graphite   | <u>0.04 Max</u>                           |
| Total  | 100.00%*                                  |

\*Data provided as listed, Total actually equals 99.98%.

**Appendix B: ANIMAL DATA**

Species: *Oryctolagus cuniculus*

Strain: New Zealand White (albino)

Source: Elkhorn Rabbitry  
5265 Starr Way  
Watsonville, CA 95076

Sex: Female

Age: Young adults

Animals in each group: 8 females

Condition of animals at start of study: Normal

Body weight range at dosing: 2.5 to 3.0 kg

Identification procedures: Ear tag, tag numbers  
85F301 - 85F308 inclusive.

Pretest conditioning:

1. Quarantine/acclimation from 14 November - 2 December 1985
2. Animal were close-clipped and examined 24 hours before dosing.

Justification: Laboratory rabbits are a proven sensitive animal model for dermal irritation studies.

**Appendix C: HISTORICAL LISTING OF STUDY EVENTS**

| <u>Date</u>          | <u>Event</u>   |
|----------------------|--|
| 14 Nov 85            | Rabbits arrived at LAIR, were examined for illness, and were placed under a 2-week quarantine.                               |
| 15 Nov 85            | Animals were weighed.  |
| 18 Nov 85            | Animals were tattooed,. All rabbits were treated with Canex <sup>®</sup> and mineral oil in their ears to prevent ear mites. |
| 14 - 27 Nov 85       | Animals were checked daily by Division of Animal Care and Services personnel.  |
| 27 Nov 85            | Rabbits were removed from quarantine after being certified healthy by a staff veterinarian. The animals were weighed.        |
| 27 Nov -<br>2 Dec 85 | Animals were checked daily.  |
| 2 Dec 85             | Animals were close-clipped and areas marked.   |
| 3 Dec 85             | Animals were weighed. Test substance was applied for 4 hours. Patches were removed and sites were scored after one hour.     |
| 3 - 17 Dec 85        | Animals were observed daily.   |
| 3 - 6 Dec 85         | Areas were scored at 1, 24, 48, and 72 hours after patch removal.  |
| 10 Dec 85            | Animals were weighed and scored.   |
| 17 Dec 85            | Animals were weighed, scored and submitted to necropsy.  |

## Appendix D: DERMAL IRRITATION DATA

| ANIMAL<br>NUMBER | OBSERVATION         | QUADRANT*   |            |            |            |
|------------------|---------------------|-------------|------------|------------|------------|
|                  |                     | I           | II         | III        | IV         |
| 85F301           | 30-60 min<br>24 hr# | 1/0†<br>0/0 | 0/0<br>0/0 | 0/0<br>0/0 | 0/0<br>0/0 |
| 85F302           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |
| 85F303           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |
| 85F304           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |
| 85F305           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |
| 85F306           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |
| 85F307           | 30-60 min<br>24 hr# | 0/0<br>0/0  | 1/1<br>0/0 | 1/1<br>0/0 | 0/0<br>0/0 |
| 85F308           | 30-60 min#          | 0/0         | 0/0        | 0/0        | 0/0        |

\* Quadrant I = sham; II, III = treated; IV = saline

† Scores are displayed as erythema/edema

# Scores were 0/0 in all quadrants for remaining observations

**Appendix D (cont.): DERMAL IRRITATION DATA**  
**(Test/Sham/Vehicle)**

| <b>ANIMAL<br/>NUMBER</b> | <b>30-60 Min</b> | <b>24 h</b> | <b>48 h</b> | <b>72 h*</b> |
|--------------------------|------------------|-------------|-------------|--------------|
| 85F301                   | 0/1/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F302                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F303                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F304                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F305                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F306                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F307                   | 1/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| 85F308                   | 0/0/0            | 0/0/0       | 0/0/0       | 0/0/0        |
| Mean                     | .125/.125/0      | 0/0/0       | 0/0/0       | 0/0/0        |
| Net Mean<br>Score†       | 0                | 0           | 0           | 0            |

\* Identical scores were recorded at 7 and 14 days.

† Test Mean - (greater of Vehicle or Sham Mean) = Net Mean Score  
 The peak net mean score is 0.0; therefore, the Primary Skin Irritation  
 Category is I (non-irritant).

Appendix E: PATHOLOGY REPORT

LAIR GLP Study 85819  
 Dermal Sensitization Study  
 Primary Investigator: CPT Earl Morgan

Compound: JA2 Propellant/Saline.

Animals: Rabbit/New Zealand White/Females/DOB: 30 August 1985.

Reference: SOP-OP-STX-34.

Procedures:

Euthanasia: Sodium pentobarbital.  
 Fixative: 10% Neutral buffered formalin.  
 Histopath: Routine

Gross findings: All animals presented live. Asterick (\*) indicates tissues saved for histopathology.

| <u>LAIR ACC#</u> | <u>ANIMAL ID#</u> | <u>OBSERVATIONS</u>  |
|------------------|-------------------|--|
| 38681            | 85F301            | Cecum - pinworms   |
| 38682            | 85F302            | Cecum - pinworms   |
| 38683            | 85F303            | Cecum - pinworms   |
| 38684            | 85F304            | Not remarkable (NR)  |
| 38685            | 85F305            | Cecum - pinworms<br>*Large - right apical lobe<br>has 1x2cm darkened focus |
| 38686            | 85F306            | Cecum - pinworms   |
| 38687            | 85F307            | Cecum - pinworms   |
| 38688            | 85F308            | Cecum - pinworms   |

Microscopic findings:

| <u>LAIR ACC#</u> | <u>ANIMAL ID#</u> | <u>MICROSCOPIC DIAGNOSES</u>  |
|------------------|-------------------|---|
| 38685            | 85F305            | Peribronchitis and peribronchiolitis;<br>lymphocytic, multifocal, mild;<br>with minimal airway ectasia. |

Comment: None of the gross or microscopic findings are compound related.

*MIV Slayter*  
 MICHAEL V. SLAYTER, DVM  
 MAJ, VC  
 C, Comparative Pathology Branch

*G. Tracy Marovec*  
 G. TRACY MAROVEC, DVM  
 MAJ, VC  
 Diplomate, ACVP  
 Comparative Pathology Branch

21 March 1986

## Distribution List

**Commander**

US Army Biomedical Research and  
Development Laboratory (12)  
ATTN: SGRD-UBZ-C  
Fort Detrick, Frederick, MD 21701-5010

Defense Technical Information Center  
(DTIC) (2)

ATTN: DTIC-DLA  
Cameron Station  
Alexandria, VA 22304-6145

US Army Medical Research and  
Development Command (2)  
ATTN: SGRD-RMI-S  
Fort Detrick, Frederick, MD 21701-5012

Commandant  
Academy of Health Sciences, US Army  
ATTN: AHS-CDM  
Fort Sam Houston, TX 78234

Chief  
USAEHA Regional Division, West  
Fitzsimmons AMC  
Aurora, CO 80045

Chief  
USAEHA Regional Division, North  
Fort George G. Meade, MD 20755

Chief  
USAEHA Regional Division, South  
Bldg. 180  
Fort McPherson, GA 30330

Commander  
USA Health Services Command  
ATTN: HSPA-P  
Fort Sam Houston, TX 78234

Commander US Army Materiel  
Command  
ATTN: AMSCG  
5001 Eisenhower Avenue  
Alexandria, VA 22333

**Commander**

US Army Environmental Hygiene  
Agency  
ATTN: Librarian, HSDH-AD-L  
Aberdeen Proving Ground, MD 21010

**Dean**

School of Medicine  
Uniformed Services University of the  
Health Sciences  
4301 Jones Bridge Road  
Bethesda, MD 20014

**Commander**

US Army Materiel Command  
ATTN: AMCEN-A  
5001 Eisenhower Avenue  
Alexandria, VA 22333

**HQDA**

ATTN: DASG-PSP-E  
Falls Church, VA 22041-3258

**HQDA**

ATTN: DAEN-RDM  
20 Massachusetts, NW  
Washington, D.C. 20314

CDR, US Army Toxic and Hazardous  
Material Agency

ATTN: DRXTH/ES  
Aberdeen Proving Ground, MD 21010

**Commandant**

Academy of Health Sciences  
United States Army  
ATTN: Chief, Environmental  
Quality Branch  
Preventive Medicine Division  
(HSHA-IPM)  
Fort Sam Houston, TX 78234