Institute Report No. 351

Acute Oral Toxicity of Ball Powder® in ICR Mice

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MAMMALIAN TOXICOLOGY BRANCH
DIVISION OF TOXICOLOGY

July 1989

Toxicology Series: 130

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

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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)*

Richard A. Kishimoto
COL, MSC
Acting Commander

24 July 1989
(date)

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The acute oral toxicity of Ball Powder® was determined in male and female ICR mice by using the oral gavage single-dose method. The median lethal dose was greater than a "limit dose" of 5000 mg/kg. Clinical signs (hypotonia, hunched posture, and diarrhea) were observed in only two of the twenty dosed animals and were minimal in both severity and duration. According to the classification scheme of Hodge and Sterner, these results place Ball Powder® in the practically nontoxic class of chemicals.

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ABSTRACT

The acute oral toxicity of Ball Powder® was determined in male and female ICR mice by using the oral gavage single-dose method. The median lethal dose was greater than a "limit dose" of 5000 mg/kg. Clinical signs (hypotonia, hunched posture, and diarrhea) were observed in only two of twenty dosed animals and were minimal in both severity and duration. According to the classification scheme of Hodge and Sterner, these results place Ball Powder® in the practically nontoxic class of chemicals.

KEY WORDS: Acute oral toxicity, Ball Powder®, Nitrocellulose, Mammalian Toxicology, Mouse, Propellant

PREFACE

TYPE REPORT: Acute Oral Toxicity 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:

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

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

GLP STUDY NUMBER: 84035

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

PRINCIPAL INVESTIGATOR: Gerald F.S. Hiatt, PhD

CO-AUTHOR: Denzil F. Frost, MS, DVM, CPT, VC
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PATHOLOGIST: George T. Makovec, DVM, MAJ, VC
Diplomate American College of
Veterinary Pathologists

REPORT AND DATA MANAGEMENT: A copy of the final report, study protocol, SOPs, raw data, analytical, stability, and purity data of the test compound, tissues, and an aliquot of the test compound will be retained in the LAIR Archives.

TEST SUBSTANCE: Ball Powder®

INCLUSIVE STUDY DATES: 27 March - 30 April 1985

OBJECTIVE: The objective of this study was to determine the acute oral toxicity of Ball Powder® in male and female ICR mice.

ACKNOWLEDGMENTS

SSG James D. Justus, SP4 James J. Fischer, and SP4 Scott L. Schwebe provided technical assistance in the research. SP4 Paul B. Simboli, SP4 John R.G. Ryabik, Richard D. Spieler, Charlotte L. Speckman, and Diane Arevalo provided care for the animals. Colleen S. Kamiyama, Ann L. Wilkinson, and Julie A. Peacock provided administrative and clerical support during the performance of this study and preparation of the report.

SIGNATURES OF PRINCIPAL SCIENTISTS AND MANAGERS

We, the undersigned, declare that GLP study number 84035 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 24 July 89

DON W. KORTE JR, PhD/DATE
MAJ, MS
Study Director

Denzil F. Frost 1 Dec 88

DENZIL F. FROST, MS, DVM/DATE
CPT, VC
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Conrad Wheeler 24 July 89

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Analytical Chemist

DEPARTMENT OF THE ARMY

LETTERMAN ARMY INSTITUTE OF RESEARCH
PRESIDIO OF SAN FRANCISCO, CALIFORNIA 94129-6800

REPLY TO
ATTENTION OF:

SGRD-ULZ-QA

24 July 1989

MEMORANDUM FOR RECORD

SUBJECT: GLP Compliance for GLP Study 84Ø35

1. This is to certify that the protocol for LAIR GLP Study 84Ø35 was reviewed on 1 November 1984.

2. The institute report entitled "Acute Oral Toxicity of Ball Powder® in ICR Mice," Toxicology Series 13Ø, was audited on 2Ø July 1989.

Carolyn M. Lewis

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

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Acute Oral Toxicity of Ball Powder® in Mice
-- Hiatt *et al.*

INTRODUCTION

Nitroguanidine, a primary component of US Army triple-base propellants, is now produced in a Government-owned contractor-operated ammunition plant. The US Army Biomedical Research and Development Laboratory (USABRDL), as part of its mission to evaluate the environmental and health hazards of military-unique pollutants generated by US Army munitions-manufacturing facilities, conducted a review of the nitroguanidine data base and identified significant gaps in the toxicity data (1). The Division of Toxicology, LAIR, was tasked by USABRDL to develop a genetic and mammalian toxicity profile for nitroguanidine, related intermediates/by-products of its manufacture, and its environmental degradation products. A genetic and acute mammalian toxicity profile of Ball Powder®, a fielded nitrocellulose-based propellant, was also requested as a baseline against which future formulations will be compared.

Objective of Study

The objective of this study was to determine the acute oral toxicity of Ball Powder® in male and female ICR mice.

MATERIALS

Test Substance

Chemical Name: Ball Powder® (Olin WC 844 double base spheroidal propellant)

Code Number: LAIR Code No. TA45

Chemical Composition:

Component	Percent
Nitroglycerin	10.235
Dinitrotoluene	0.685
Diphenylamine	1.105
Dibutylphthalate	5.255
Nitrocellulose	83.23
Total Volatiles	1.045
Moisture and Volatiles	0.895
Residual Solvent	0.49
Calcium Carbonate	0.09
Sodium Sulfate	0.12

Source: Badger Army Ammunition Plant
Baraboo, WI 53913

Other information on the test substance is presented in Appendix A.

Vehicle

The vehicle for Ball Powder® was 1% carboxymethylcellulose (Sigma Chemical Co, St Louis, MO), expiration 1 July 1991, in sterile water for injection (Abbott Laboratories, North Chicago, IL).

Animal Data

Twenty male and 20 female ICR mice (Harlan Sprague Dawley, Inc., Indianapolis, IN) from a shipment that arrived on 27 Mar 84 were used in this study. They were identified individually with cervical tags numbered 85C00325 to 85C00344 (males) and 85C00345 to 85C00364 (females), inclusive. Two males and two females were selected for quality control necropsy evaluation at receipt. The animal weights on 27 Mar 85 ranged from 20 to 33 g. Additional animal data appear in Appendix B.

Husbandry

Mice were maintained individually in stainless steel wire mesh cages in racks equipped with automatically flushing dump tanks. No bedding was used in any of the cages. The diet, fed *ad libitum*, consisted of Certified Purina Rodent Chow® Diet 5002 (Ralston Purina Company, Checkerboard Square, St Louis, MO); water was provided by continuous drip from a central line. The temperature of the animal room was maintained at a range from 20° C to 28.3° C and a relative humidity range of 20 to 58%; with temporary spikes to 64% during room washing. The photoperiod was 12 hours of light per day.

METHODS

Group Assignment/Acclimation

Study mice were assigned to a dose group of 10 males and 10 females and a vehicle control group of 5 males and 5 females each. The animals were acclimated for 18 days before the day of dosing. During this period they were observed daily for signs of illness.

Dose Levels

Since the median lethal dose (MLD) of the major component of Ball Powder®, nitrocellulose, is greater than 5000 mg/kg (2), a "limit dose" of 5000 mg/kg was selected for evaluating the acute oral toxicity of Ball Powder®.

Preparation of Compound

The Ball Powder® was ground in a freezer mill under liquid nitrogen, sieved through a 100-mesh screen, and then suspended in the vehicle with a spatula. The carboxymethylcellulose (CMC) vehicle had been prepared by mixing 1 g of CMC in 100 ml of sterile water using a Kinematica model CH-6010 homogenizer.

Chemical Analysis of Dosing Suspension

Since Ball Powder® is a complex mixture, analysis of the dosing solution for accuracy and stability was not technically feasible. To ensure the accuracy of the administered dose, the suspension was prepared immediately before administration. The entire procedure was completed within 30 minutes.

Test Procedures

This study was conducted in accordance with EPA guidelines (3) and LAIR SOP-OP-STX-36, (4).

The volume of dosing suspension each animal received was based upon the desired dose level, the concentration of the compound in the suspension, and the weight of the animal. The dosing volume for all groups was based on a standard of 10 ml/kg per animal. Volumes ranged from 0.28 to 0.36 ml in the males and from 0.28 to 0.34 ml in the females. The vehicle control group for males and females was 0.32 to 0.37 ml and 0.28 to 0.33 ml, respectively, of a 1% carboxymethylcellulose suspension. Dosing was performed using the oral gavage method without sedating the animal or administering anesthesia. Sterile disposable syringes (Becton, Dickerson & Co, Rutherford, NJ) fitted with 18-gauge, 3-inch, ball-tipped feeding tubes (Popper & Sons, Inc., New Hyde Park, NY) were used for dosing. The test compound and vehicle animals were dosed between 1044 and 1142 hours on 16 April 1985.

Observations

Observations for mortality and signs of acute toxicity were performed daily according to the following procedure: (a) animals were observed undisturbed in their cages, (b) animals were removed from their cages and given a physical examination, and (c) animals were observed after being returned to their cages. On the day of dosing, the animals were checked intermittently throughout the day. Recorded observations were performed 2 and 4 hours after dosing, and daily for the remainder of the 2-week test period. A second "walk-through" observation was performed daily and only significant observations were recorded. Body weights were recorded once weekly during the course of the study.

Necropsy

All animals were submitted for a complete gross necropsy immediately after receiving a barbiturate overdose.

Duration of Study

Appendix C is a complete listing of historical events.

Changes/Deviations

The study was accomplished according to the protocol andapplicable amendments with the following exceptions:

On the initial dosing date (4 Apr 85), the viscosity of the test compound was such that it could not be passed through a syringe. Therefore, dosing was postponed until the viscosity could be reduced, which entailed a finer grinding of the Ball Powder®. Because of this delay, the scheduled weighing on 11 Apr 85 was inadvertently omitted. On 27 Apr 85, the exhaust fan was inoperable for 12 hours which caused the relative humidity to rise to 80%-95% with no change in temperature for that period.

Raw Data and Final Report Storage

A copy of the final report, study protocol, raw data, retired SOPs and an aliquot of the test compound will be retained in the Letterman Army Institute of Research Archives.

RESULTS

Mortality

No deaths occurred in either the test compound or the vehicle control groups.

Clinical Observations

Ten male and ten female animals were administered the limit dose of Ball Powder®. Eighteen out of twenty animals dosed appeared normal throughout the observation study. The two remaining animals exhibited hypotonia and hunched posture or orange material on the tail (diarrhea), but had returned to normal within twenty-four hours. All animals survived until termination of the study.

Table 1 contains a summary of clinical observations. Appendix D contains individual animal histories.

Weight gains of survivors were not affected by dosing. Table 2 presents the mean body weights by groups. Appendix E contains individual weight tables.

Gross Pathological Observations

No lesions were found at necropsy which could be attributed to the test compound or the test procedure. The veterinary pathologist's report appears in Appendix F.

TABLE 1: Incidence Summary for Clinical Observations in Mice Administered a Limit Dose of Ball Powder®

Category of Clinical Signs	Group Dose (mg/kg) (N=)	1 Vehicle 5	2 5000 (limit) 10
MALES			
Hypotonia		0	1
Skin[a]		1	0
Hunched Posture		0	1
Normal		4	9
FEMALES			
Miscellaneous[b]		0	1
Normal		5	9

[a] Includes scab on neck.

[b] Includes orange material on tail.

TABLE 2: Mean Body Weights for Mice Administered a Limit Dose of Ball Powder®†

Group	Receipt	Dosing Day	Day 7	Day 14
		MALES		
5000 mg/kg	28.1 ±2.6(10)	32.0 ±2.5(10)	33.9 ±1.7(10)	34.5 ±1.7(10)
Vehicle Control	27.2 ±4.3(5)	33.2 ±0.8(5)	34.8 ±0.5(5)	34.2 ±3.6(5)
		FEMALES		
5000 mg/kg	28.5 ±1.5(10)	30.3 ±2.0(10)	31.3 ±1.6(10)	31.9 ±1.5(10)
Vehicle Control	27.8 ±3.8(5)	29.6 ±2.1(5)	31.2 ±2.8(5)	33.2 ±2.3(5)

† Values are mean in g ± SD (n).

DISCUSSION

The MLD for Ball Powder® is greater than 5000 mg/kg in male and female ICR mice. This places Ball Powder® in the "practically non-toxic" class (5).

No definitive clinical syndrome or target organ/system was apparent from the recorded observations. The clinical signs observed (i.e., diarrhea, hunched posture, and hypotonia) were non-specific and could not be attributed to an effect on a particular organ system. The lack of a toxic response to Ball Powder® was surprising because even though Ball Powder® contains 83% nitrocellulose, by weight which is essentially nontoxic (2), it does contain 10.2% nitroglycerin. Nitroglycerin has appreciable toxicity--an oral MLD of 500 mg/kg in mice (6). Since the calculated amount of nitroglycerin administered in Ball Powder® to the animals in the study was 510 mg/kg, one would expect the mice to have exhibited appreciable nitroglycerin-related toxicity. The fact that this was not observed suggests that nitroglycerin is complexed in the Ball Powder® formulations in such a way that it is not readily absorbed.

CONCLUSION

Ball Powder® is a practically nontoxic compound since it produced no significant observable effects or deaths at the "limit dose" of 5000 mg/kg in male and female ICR mice.

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3. Environmental Protection Agency. Office of Pesticides and Toxic Substances, Office of Toxic Substances (TS-792). Acute exposure, oral toxicity. In: Health effects test guidelines. Washington, DC: Environmental Protection Agency, August 1982; EPA 560/6-82- 0011.

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5. Hodge HC, Sterner JH. Tabulation of toxicity classes. Amer.Ind. Hyg. Assoc. Q. 1943; 10:93-96.

6. Tatken RL, Lewis RJ, eds. Nitroglycerin QX2100000. Registry of Toxic Effects of Chemical Substances, 1981-82; Vol. 2, D-O.

Appendix A: CHEMICAL DATA

PROPELLANT DESCRIPTION SHEET

REPORTS CONTROL SYMBOL
EXEMPT - PARA 7-2a
AR 335-15

TO	FROM	DATE
	Badger Army Ammunition Plant Baraboo, Wisconsin 53913	10 August 1984

DA LOT NUMBER 50/50 blend of lots BAJ-47670 and BAJ-47671	COMPOSITION NUMBER WC 844 for Cartridge 5.56 mm, BALL, M193
MFG AT Badger Army Ammunition Plant	PACKED AMOUNT LB
CONTRACT NUMBER DAAA09-73-C-0004	SPECIFICATION NUMBER MIL-P-3984E w/Amendment 4 and Drawing No. C10542743 Rev. C

NITROCELLULOSE

ACCEPTED BLEND NUMBERS	NITROGEN CONTENT	KI STARCH(65.5°C)	STABILITY (134.5°C)
Nitrocellulose (NC) extracted from excessed	MAX %	MIN	MIN
Single Base Propellant.	MIN %	MIN	MIN
	AVG %	MIN	MIN
NC complied with MIL-N-244A			EXPLOSION HR

MANUFACTURE OF PROPELLANT

_____POUNDS SOLVENT PER POUND NC/DRY WEIGHT INGREDIENTS CONSISTING OF _____ POUNDS ALCOHOL AND _____ POUNDS _____ PER 100 POUNDS SOLVENT. PERCENTAGE REMIX TO WHOLE_____.

TEMPERATURE FROM	TEMPERATURE TO	PROCESS--SOLVENT RECOVERY AND DRYING	TIME DAYS	TIME HOURS

PROPELLANT COMPOSITION — TESTS OF FINISHED PROPELLANT — STABILITY AND PHYSICAL TESTS

CONSTITUENT	% FORMULA	% TOLERANCE	% MEASURED		FORMULA	ACTUAL
Nitroglycerin			10.235	HEAT TEST 120°	Min 60 min.	65 min.*
Dinitrotoluene			0.685	No Explosion (HRS)	Min 5	5+*
Diphenylamine			1.105	FORM OF PROPELLANT		
Dibutylphthalate			5.255	Dust&Foreign Mat.		0.02
Nitrocellulose			83.23	Graphite		0.075
Total Volatiles			1.045	Grav. Density		1.008
Moisture and Volatiles			0.895	Nitrogen		13.075
Residual Solvent			0.49			
Calcium Carbonate			0.09			
Sodium Sulfate			0.12			

CLOSED BOMB	LOT NUMBER	TEMP °F	RELATIVE QUICKNESS	RELATIVE FORCE
TEST				
STANDARD			100.00%	100.00%

PROPELLANT DIMENSIONS (INCHES)	SPEC	DIE	FINISHED	MEAN VARIATION IN % OF MEAN DIMENSIONS: SPEC	ACTUAL
LENGTH (L)					
DIAMETER (D)					
PERF DIA (d)					
				PACKED	
				SAMPLED	
WEB DIFFERENCE/STD DEV IN % OF WEB AVERAGE				TEST FINISHED	
				OFFERED	
L:D				DESCRIPTION SHEETS FORWARDED	
D:d					

REMARKS

TYPE OF PACKING CONTAINER

REMARKS
*Tested 29 February 1984.

SIGNATURE OF CONTRACTOR'S REPRESENTATIVE	SIGNATURE OF GOVERNMENT QUALITY ASSURANCE REPRESENTATIVE

Appendix B: ANIMAL DATA

Species: *Mus musculus*

Strain: ICR

Source: Harlan Sprague Dawley, Inc.
Indianapolis, IN

Sex: Male and female.

Date of birth: Male: 8 February 1985
Female: 1 February 1985

Method of randomization: According to animal number based on random numbers generated by a HP calculator.

Animals in each group: 10 male and 10 female animals
5 each for vehicle control group

Condition of animals at start of study: Normal

Body weight range at dosing: 27-36 g

Identification procedures: Cervical tag, tag numbers between 85C00325 to 85C00344 and 85C00345 to 85C00364 inclusive.

Pretest conditioning: Quarantine/acclimation 27 March 1985 - 15 April 1985.

Justification: The laboratory mouse has proven to be a sensitive and reliable system for lethal dose determination.

Appendix C: HISTORICAL LISTING OF STUDY EVENTS

Date	Event
27 Mar 85	Mice arrived and were checked for physical condition, sexed, and individually caged.
28 Mar 85	All animals were weighed, cervically tagged, and assigned to dose groups. Two males and two females were submitted for necropsy control.
28 Mar - 3 Apr 85	Animals were observed daily.
1,3 Apr 85	Animals were weighed.
4 Apr 85	Food was removed from the test animals at 0600 hours. Test compound animals were weighed, and dosing was initiated at 1100. Only one animal dosed due to excessive viscosity of the dosing suspension.
5 Apr 85	The single dosed animal was terminated.
5-15 Apr 85	All animals were observed in a.m. and p.m.
16 Apr 85	Food was removed from all test animals at 0600 hours. Animals were weighed and dosing was initiated at 1044 hours and concluded at 1142 hours. All animals were observed at 2 and 4 hours after dosing.
17-29 Apr 85	All animals were observed daily in a.m. and p.m.
23 Apr 85	All animals were weighed.
27 Apr 85	Exhaust fan was inoperable for 12 hours.
30 Apr 85	Food was removed from animals at 0600 hours. All animals were observed, weighed, and submitted to necropsy.

Appendix D: INDIVIDUAL ANIMAL HISTORIES

MALES: 5000 mg/kg Ball Powder®

Animal Number	Clinical Signs	Dates Observed (1984)	Severity
84C00327	Normal	N/A	N/A
84C00328	Normal	N/A	N/A
84C00330	Normal	N/A	N/A
84C00334	Hunched Posture Hypotonia	Apr. 16 Apr. 16	Slight Slight
84C00335	Normal	N/A	N/A
84C00336	Normal	N/A	N/A
84C00337	Normal	N/A	N/A
84C00338	Normal	N/A	N/A
84C00340	Normal	N/A	N/A
84C00341	Normal	N/A	N/A

APPENDIX D (cont.): INDIVIDUAL ANIMAL HISTORIES

FEMALES: 5000 mg/kg Ball Powder®

Animal Number	Clinical Signs	Dates Observed (1984)	Severity
84C00346	Normal	N/A	N/A
84C00347	Normal	N/A	N/A
84C00351	Normal	N/A	N/A
84C00352	Material, Orange, Tail	Apr. 16	Slight
84C00356	Normal	N/A	N/A
84C00357	Normal	N/A	N/A
84C00358	Normal	N/A	N/A
84C00360	Normal	N/A	N/A
84C00361	Normal	N/A	N/A
84C00363	Normal	N/A	N/A

Appendix D (cont.): INDIVIDUAL ANIMAL HISTORIES

Vehicle Control

Animal Number	Clinical Signs	Dates Observed (1984)	Severity
	MALES		
84C00325	Normal	N/A	N/A
84C00331	Scab, Neck	Apr. 16,17	Present
84C00332	Normal	N/A	N/A
84C00343	Normal	N/A	N/A
84C00344	Normal	N/A	N/A
	FEMALES		
84C00345	Normal	N/A	N/A
84C00349	Normal	N/A	N/A
84C00354	Normal	N/A	N/A
84C00355	Normal	N/A	N/A
84C00364	Normal	N/A	N/A

Appendix E: INDIVIDUAL BODY WEIGHTS IN GRAMS

Males: 5000 mg/kg

Animal No.	At Receipt	At Dosing	Day 7	Termination Day 14
85C00327	24*	34	34	34
85C00328	31	36	37	37
85C00330	32	35	36	37
85C00334	29	31	34	33
85C00335	29	33	34	35
85C00336	25	30	33	34
85C00337	29	30	33	35
85C00338	27	31	33	35
85C00340	29	32	34	33
85C00341	26	28	31	32
Mean	28.1	32.0	33.9	34.5
Standard Deviation	2.6	2.5	1.7	1.7
Std. Error of the Mean	0.8	0.8	0.5	0.5

* Values are given in grams.

Appendix E (cont.): INDIVIDUAL BODY WEIGHTS IN GRAMS

Females: 5000 mg/kg

Animal No.	At Receipt	At Dosing	Day 7	Termination Day 14
85C00346	26*	28	29	31
85C00347	27	28	29	30
85C00351	29	34	34	34
85C00352	31	31	32	33
85C00356	29	30	32	33
85C00357	29	31	32	32
85C00358	28	29	31	32
85C00360	29	30	31	32
85C00361	27	29	30	29
85C00363	30	33	33	33
Mean	28.5	30.3	31.3	31.9
Standard Deviation	1.5	2.0	1.6	1.5
Std. Error of the Mean	0.5	0.6	0.5	0.5

* Values are given in grams.

Appendix E (cont.): INDIVIDUAL BODY WEIGHTS IN GRAMS

Vehicle Control

Animal No.	At Receipt	At Dosing	Day 7	Termination Day 14
		MALES		
85C00325	29*	33	34	34
85C00331	31	34	35	37
85C00332	29	34	35	33
85C00343	20	33	35	29
85C00344	27	32	35	38
Mean	27.2	33.2	34.8	34.2
Standard Deviation	4.3	0.8	0.5	3.6
Std. Error of the Mean	1.9	0.4	0.2	1.6
		FEMALES		
85C00345	22	28	28	31
85C00349	27	28	30	33
85C00354	30	30	33	33
85C00355	32	33	35	37
85C00364	28	29	30	32
Mean	27.8	29.6	31.2	33.2
Standard Deviation	3.8	2.1	2.8	2.3
Std. Error of the Mean	1.7	0.9	1.2	1.0

* Values are given in grams.

Appendix F: PATHOLOGY REPORT

LAIR Gross Pathology Report
GLP Study 84035

Study: GLP #84035, Toxicology Services Group

Test: Oral Acute Toxicity (LD_{50}) Limit Test.

Investigator: Dr. Gerald Hiatt.

Test Substance: Ball powder.

History: Study conducted in accordance with SOP-OP-STX-36. Number of animals: 30 Sex: 15 male, 15 female Species: Mus musculus Breed: ICR. GLP Study #84035.

Gross findings:

Dosage Level	Sex	No. of animals	Deaths	Lesions
5,000 mg/kg	M	10	0	1*
Vehicle	M	5	0	0
5,000 mg/kg	F	10	0	0
Vehicle	F	5	0	0

*Mouse number 85C00327 (LAIR ACC # 37501) had a dark red 3 x 4 mm diameter focal area on the left lateral lobe of the liver. Microscopically there were several random small foci of acute coagulative necrosis of hepatocytes with pooling of blood in sinusoids and mild infiltrates of neutrophils and macrophages. (Hepatitis, necrotizing, acute, multifocal, mild, liver.) Special stains were noncontributory.

Comments: The liver lesion was considered not to be directly related to the test compound. The cause of the liver necrosis is not known.

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