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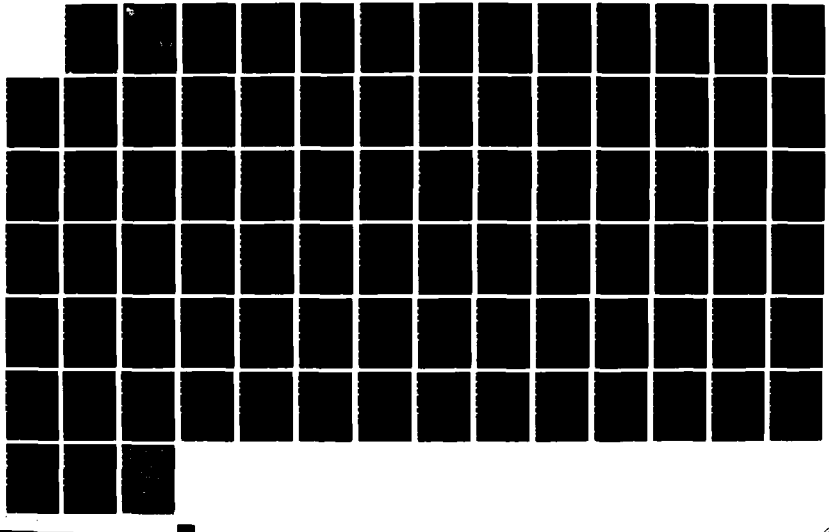

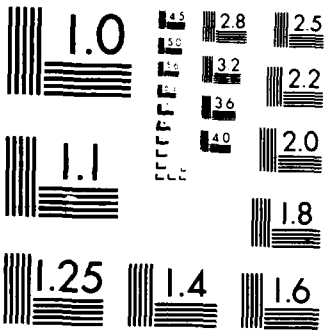
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Institute Report No. 255

Fourteen-Day Subchronic Oral Toxicity Study of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate in Male Rats

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Mammalian Toxicology Branch
Division of Toxicology

February 1988

Toxicology Series: 74

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

Fourteen Day Subchronic Oral Toxicity Study of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate in Male Rats (Toxicology Series 74)--
Lewis, White, Kellner, Waring, Turnier, and Fruin

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

The 14-day subchronic oral toxicity of 4-nitrophenyl monochloromethyl (phenyl phosphinate (MCP) was evaluated in male rats. MCP was administered by gavage at dose levels of 0, 12.5, 25, 50 and 100 mg/kg/day for 14 days. At necropsy blood samples were obtained for hematological and serum clinical analyses. A complete histological examination was performed on all animals. In addition, plasma, red blood cell, and brain acetylcholinesterase and butyrylcholinesterase activities were determined. Although MCP was lethal to one rat in both the 50 and 100 mg/kg dose groups, no definitive pattern of clinical chemical, hematological or histopathological alterations was found. This suggests that the deaths observed could be due to a transient toxic response associated with cholinesterase inhibition.

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ABSTRACT

The 14-day subchronic oral toxicity of 4-nitrophenyl monochloromethyl (phenyl) phosphinate (MCP) was evaluated in male rats. MCP was administered by gavage at dose levels of 0, 12.5, 25, 50 and 100 mg/kg/day for 14 days. At necropsy, blood samples were obtained for hematological and serum clinical analyses. A complete histological examination was performed on all animals. In addition, plasma, red blood cell, and brain acetylcholinesterase and butyrylcholinesterase activities were determined. Although MCP was lethal to one rat in both the 50 and 100 mg/kg dose groups, no definitive pattern of clinical chemical, hematological or histopathological alterations was found. This suggests that the deaths observed could be due to a transient toxic response associated with cholinesterase inhibition.

Key Words: Subchronic Oral Toxicity, 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate, Phosphinates, Rat

PREFACE

TYPE REPORT: Fourteen-Day Subchronic 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: US Army Medical Research and Development Command
US Army Medical Research Institute of Chemical Defense
Aberdeen Proving Ground, MD 21010-5425

PROJECT/WORK UNIT/APC: 35162772A875 Defense Against Chemical Agents, WU 304, Toxicity Testing of Phosphinate Compounds, APC TL04

GLP STUDY NUMBER: 82034

STUDY DIRECTOR: COL John T. Fruin, DVM, PhD, VC,
Diplomate, American College of
Veterinary Preventive Medicine

PRINCIPAL INVESTIGATORS: CPT Craig W. White, DVM, VC
Carolyn M. Lewis, MS
SP5 Thomas P. Kellner, BA

PATHOLOGIST: John C. Turnier, DVM, MAJ, 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, tissues, and an aliquot of the test compound will be retained in the LAIR Archives.

TEST SUBSTANCE: 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

INCLUSIVE STUDY DATES: 17 November - 16 December 1982

OBJECTIVE: The objective of this study was to determine the subchronic toxicity of 4-nitrophenyl monochloromethyl (phenyl) phosphinate (MCP) in male rats.

ACKNOWLEDGMENTS

SP5 L. Sauers, MS; SP5 L. Mullen, BS; SP5 J. Rodriguez, BS; and SP5 E. Zimmerman assisted with daily dosing and observations. CPT(P) G. Makovec, DVM; CPT(P) M. Langford, DVM; SSG C. Beckett; SP5 M. McKinley, BA; SP5 F. McKinley, BA; SP5 T. Loughead; SP4 C. Dumlao, BS; SP4 M. Kostrna; L. Cote and T. Hironaga contributed in the collection, preparation and histological examination of tissues and in performing the hematology and urinalysis. M. Lyons and J. Knudsen, BS, performed the various biochemical analyses. Claire N. Lieske, US Army Research Institute of Chemical Defense, provided the compound, advice, and support.

SIGNATURES OF PRINCIPAL SCIENTISTS AND MANAGERS INVOLVED IN THE STUDY:

We, the undersigned, declare that GLP study number 82034 was performed under our supervision, according to the procedures described herein, and that this report is an accurate record of the results obtained.

JOHN T. FRUIN / DATE
COL, VC
Study Director

THOMAS P. KELLNER, BS / DATE
SP5, USA
Principal Investigator

JOHN C. TURNIER / DATE
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DEPARTMENT OF THE ARMY

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

REPLY TO
ATTENTION OF:

SGRD-ULZ-QA (70-1n) 13 January 1988

MEMORANDUM FOR RECORD

SUBJECT: Report of GLP Compliance for Study 82034

1. I hereby certify that in relation to LAIR GLP Study 82034, the following inspections were made:

02 November 1982 - Protocol Review
03 December 1982 - Dose Preparation
03 December 1982 - Dosing
14 December 1982 - Observations
15 December 1982 - Necropsy
15 December 1982 - Tissue Processing
15 December 1982 - Clinical Chemistry

2. The report and raw data for this study were audited on 26 May 1987.

GARY L. DUTCHER
Principal Advisor
Quality Assurance Section

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APPENDICES

FOURTEEN-DAY SUBCHRONIC ORAL TOXICITY STUDY OF 4-NITROPHENYL MONOCHLOROMETHYL (PHENYL) PHOSPHINATE IN MALE RATS--Lewis et al

One mission of the US Army Medical Research and Development Command is to develop a prophylactic regimen against organophosphate intoxication. The organophosphinate compounds offer an effective strategy of prophylaxis. The strategy requires protecting a critical percentage of the available acetylcholinesterase from irreversible binding during chemical agent poisoning. This is accomplished by reversible binding with a compound, such as 4-nitrophenyl monochloromethyl (phenyl) phosphinate, from which the enzyme may be reactivated using standard antidotal therapy (1-4).

Objective of the Study

The objective of this study was to determine the subchronic toxicity of 4-nitrophenyl monochloromethyl (phenyl) phosphinate (MCP) in male rats.

MATERIALS

Test Substance

Chemical name: 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

LAIR Code: TA009

Code name: MCP, CMP

Chemical Abstract Service Registry Number: None

Empirical formula: $C_{13}H_{11}ClNO_4P$

The test compound was received from the US Army Medical Institute of Chemical Defense, Aberdeen Proving Ground, MD 21010 on 23 June 1982. The test chemical was stored at 4°C until the time of compounding with the vehicle before dosing. Detailed chemical data on the test compound are given in

Appendix A.

Vehicle

The vehicle contained 20% Tween 80™ (Fisher Scientific Company, Fairlawn, NJ), 10% ethanol and 70% citrate buffer (pH 3.0). This vehicle was selected because it significantly retarded phosphinate hydrolysis.

Animals

Sixty-eight male albino Sprague-Dawley rats were received from Bantin-Kingman Breeding Laboratories, Fremont, CA for use in this study. Ear tags, numbers 82D00974 to 82D01041, without exclusions, were used to identify each animal individually. Two animals were sacrificed for quality control necropsies, six extra animals were eliminated during randomization as extras, and two other animals were removed from the study after being misdosed. The rats' weights (17 November 1982) ranged from 145 to 177 g.

Husbandry

The animals in this study were housed individually in stainless steel mesh drawer rack cages. No bedding was used in any of the cages.

Diet consisted of Certified Purina Rodent Chow No. 5002 (Ralston Purina, Checkerboard Square, St. Louis, MO, Lot No. OCT14822F and SEPT09822K) ad libitum. Water was provided by automatic Lixit dispenser.

The temperature range maintained throughout this study was 20-26°C with a relative humidity of 40-55% with occasional spikes up to 72% during room cleaning. The photoperiod was 15 hours of light daily (0500-2000 hours).

METHODS

Group Assignment/Acclimation

The animals were acclimated for 13-14 days from receipt to the day of dosing. During the acclimation period the animals were observed daily for signs of illness.

Ten male animals were assigned to each of six dose groups. Allocation was accomplished using a computer-based stratified, randomization method (LAIR SOP-OP-STX-78).

Dose Levels

The dose for each animal was based on the body weight and the assigned dose group. Doses were calculated by a program on a Hewlett-Packard 98A calculator (LAIR SOP OP-ISG-8). The animals were weighed twice a week and doses were adjusted accordingly. The volume administered ranged from 0.21 to 2.7 ml depending on dosage and animal weights.

Four dose levels were given to male rats (10 animals/dose level) at 1/16, 1/8, 1/4, and 1/2 of the acute LD_{50} for MCP (200 mg/kg) each day. Table 1 in Appendix B shows the dosing scheme. Each dose group was further divided into two subgroups. One subgroup (a) was dosed beginning on 1 Dec 82 and the other subgroup (b) on 2 Dec 82. This procedure reduced the number of animals sacrificed on one day to a manageable level.

Compound Preparation

The solutions for the vehicle control group were prepared just before the study started. The MCP dosing solutions were prepared daily according to LAIR SOP OP-STX-48, "Preparation of Phosphinate Compounds for Oral Toxicity Studies", except that the concentrations of Tween 80", ethanol, water, and citrate buffer were changed to minimize hydrolysis. The dosing solutions were analyzed for hydrolysis (stability) immediately after preparation and within 20 minutes after dosing was completed. The results from these analyses are given in Appendix A.

Test Procedures

All animals were dosed daily between 0830 and 1030 hours for 14 days. The animals were not fasted. An 18-gauge, 3-inch gastric gavage needle (Popper and Sons, Inc., New Hyde Park, NY 11040) was used to administer the compound by gastric intubation. This procedure was performed without administering sedatives or anesthesia to the animals.

One hour after each dosing the animals were observed for mortality and signs of toxicity. Animals were observed undisturbed in cages, outside of cages, and after return to cages. If an animal exhibited severe signs of toxicity, it was observed more frequently. Moribund animals were euthanized and submitted for necropsy. Body weights were recorded twice weekly and on the day of sacrifice. Appendix C contains a listing of the historical events.

All animals assigned to this study were subjected to complete necropsy procedures. All tissues itemized in SOP OP-

STX-52 were examined microscopically in the cage control, vehicle control, and high dose groups. The other three dose groups had histopathology performed only on the liver, kidney, heart, and those organs with gross lesions. Hematology and blood chemistry analyses were also performed. A list of LAIR SOPs used for the blood chemistry is in Appendix D.

Changes to the original protocol are discussed in Appendix E.

Statistics

The animal weights and the results from hematology and blood chemistry analyses were analyzed statistically with packaged programs available on BMDP software (5). The equality of the variances of the groups was tested using the Levene's Test. If the variances were equal, the vehicle control group and the dose groups were compared by the standard one-way analysis of variance (ANOVA). Otherwise, the Welch one-way ANOVA, which is not based on the assumption that the variances are equal, was performed. If the F-statistic was significant in either case, the Dunnett's test was performed to determine whether or not the vehicle control group was significantly different from any of the dose groups. The Student's t-test was used to compare all values of the cage and vehicle control groups except total bilirubin. If the variances of the two control groups were not equal by the Levene's test, the t-statistic was calculated with the variance of each group estimated separately; otherwise, it was calculated with the variances pooled (averaged). Total bilirubin values were nonparametric data which were analyzed by using the Kruskal-Wallis one-way ANOVA. The total bilirubin levels in the two control groups were compared by using the Mann-Whitney test.

RESULTS

Mortalities

Four deaths were observed during the study; however, two of the four mortalities were attributed to misdosing. Animals 82D01009 (12.5 mg/kg group) and 82D01019 (100 mg/kg group) were removed from the study based on the pathology report. The other two deaths, one at 50 mg/kg and one at 100 mg/kg, were compound-related (Table 1, Appendix B).

Clinical Signs

MCP produced dose-dependent increases in the incidence rate of some signs. These signs included sluggishness or inactivity, excitation, decreased respiratory rate, rough

coat, excessive salivation (clear or yellow material around the mouth and on the front legs), yellow stain/material around the perianal and ventral areas (presumably urine), and red stain/material around the mouth, nose, head and neck (presumably harderian gland secretions).

A few signs were seen less frequently, but did occur primarily in the higher dose groups suggesting that they were more severe signs of toxicity. These included aggressiveness, loss of equilibrium, increased respiratory rate, increased or decreased respiratory depth, wheezing, hunched posture, orange or clear stain perianal, and brown urine.

Individual clinical signs appear in Appendix F-1.

Animal Weights

The mean body weights and standard error of the mean for each group are given in Table 2, Appendix B. The body weights for the vehicle control and test groups were not significantly different when compared by ANOVA. When the control groups were compared, the vehicle control group had significantly lower weights than the cage control group on the last three weighings.

Individual body weights appear in Appendix F-2.

Clinical Chemistry

The effect of MCP on the level of several electrolytes, various biochemical components, and the activity of several enzymes in serum was examined. In addition, acetylcholinesterase and butyrylcholinesterase activity were analyzed in plasma, red blood cells, and brain tissue. The mean and standard error of the mean for each dose group for these measurements are shown in Tables 3 through 6, Appendix B.

When the vehicle control and dose groups were compared by ANOVA, significant differences were found with the levels of blood urea nitrogen, creatine phosphokinase, and alkaline phosphatase in serum and acetylcholinesterase in brain. However, when the Dunnett's test was performed, no significant differences were found except with alkaline phosphatase levels. The high dose group (100 mg/kg/day) had significantly lower alkaline phosphatase levels than the vehicle control group. When the vehicle and cage control groups were compared using the Student's *t*-test, no differences were found in any clinical chemistry values.

Individual clinical chemistry values appear in Appendix F-3.

Pathology/Hematology

Gross necropsies of the two rats whose deaths were attributed to the compound revealed signs of gastric irritation. Gross necropsy findings in terminally sacrificed rats include dilated renal pelvis in one vehicle control and one 100 mg/kg rat, thickening of the splenic capsule in one 12.5 mg/kg rat, a focal skin abrasion in another 12.5 mg/kg rat, and yellow-brown and red-brown pulmonary foci in one 25 mg/kg and one 50 mg/kg rat, respectively.

The histopathological lesions found in terminally sacrificed rats included peritracheal hemorrhage in one 100 mg/kg rat, periesophagitis in two 100 mg/kg rats, interstitial pneumonitis in two vehicle control, one 50 mg/kg and two 100 mg/kg rats, hemorrhage and/or erythrophagocytosis in the mesenteric lymph nodes of four 100 mg/kg rats, portally oriented subacute hepatitis in two 50 mg/kg and three 100 mg/kg rats, and renal tubular mineralization in one cage control rat, one vehicle control rat, six 12.5 mg/kg rats, six 25 mg/kg rats, three 50 mg/kg rats and three 100 mg/kg rats. The histopathological findings in the two rats that died from the compound included renal tubular mineralization in the 100 mg/kg rat, periportal subacute hepatitis in both rats, hepatic necrosis in the 50 mg/kg rat, hemorrhage and/or erythrophagocytosis in the mesenteric lymph node of the 100 mg/kg rat, acute gastric inflammation in the 50 mg/kg rat, gastric hemorrhage in both rats, slight intestinal hemorrhage in the 100 mg/kg rat and slight necrosis of the stomach and intestines in the 50 mg/kg rat.

The effect of MCP on various hematological measurements was examined. The mean and standard error of the mean for each group are shown in Table 7, Appendix B. When the control groups were compared by the Student's t-test, no significant differences were found in any of the measurements. When the dose groups and the vehicle control group were compared by ANOVA, a few significant differences were found. The 12.5 mg/kg group had significantly higher hematocrits than the vehicle control group. The mean corpuscular hemoglobin values were significantly lower in the 50 mg/kg dose group than the vehicle control group. In addition, the mean corpuscular hemoglobin concentration values in the 100 mg/kg dose group were significantly lower than the vehicle control group.

The pathology report appears in Appendix G-1. Individual hematology values appear in Appendix G-2.

DISCUSSION

The types of clinical signs observed in the 14-day subchronic study of MCP were similar to those reported in the acute study (6), although the frequency and the severity were usually lower. Nearly all the signs observed could be attributed to effects of MCP on the nervous system. The most frequent signs were sluggishness or inactivity, excitation, loss of equilibrium, changes in respiration, excessive salivation (often yellow presumably from hydrolysis of the compound), excessive urination, excessive harderian gland secretions and piloerection.

Although the body weights for the vehicle control group were not significantly different from those of the test groups at any time during the study, they were significantly lower than the cage control group after the first week of dosing. There are several possible explanations for their lower weights. The animals may have been traumatized by the dosing which affected their appetite, or the vehicle itself may have affected their appetite. The vehicle could have also affected the absorption or transit time so that less food was absorbed. At this point we cannot be certain which, if any, of these factors contributed to the weight differences observed in this study.

A few statistically significant differences were seen in the clinical chemistry data. Of these few differences, none appeared to be compound-related. Alkaline phosphatase levels were significantly lower in the highest dose group when compared to the vehicle control group. In general, one is concerned about elevated levels of alkaline phosphatase, not decreased levels. This difference was considered incidental.

The difference in the creatine phosphokinase levels between groups was significant when the Welch one-way ANOVA was performed. However, none of the treatment groups were significantly different from the vehicle control group by the Dunnett's test. The difference found with the ANOVA was due primarily to elevated levels in two animals in the 50 mg/kg/day dose group. Creatine phosphokinase is particularly sensitive to skeletal muscle damage. Even exercise, intramuscular injections, and psychotic reactions can result in elevated levels (7). Since MCP is known to cause tremors, convulsions, and fasciculation, elevated levels in a few animals are not surprising. One of these animals had slight to moderate signs of toxicity the day before sacrifice; however, the other animal never exhibited any signs of toxicity during the study period. The pathology report and other clinical chemistry results were examined for these two animals, but no other evidence supporting the possibility of

muscle damage was found.

Pathological examinations of the rats that died and those that survived the 14-day dosing period revealed few distinct compound-related effects. Two rats that appeared to have died from the compound exhibited signs of gastrointestinal irritation. Gross necropsy findings in the rats that survived were regarded as minimal and considered unrelated to the compound administration. Microscopic examination of these rats revealed hemorrhages within the lymph nodes, portally oriented hepatic inflammation and renal tubular mineralization in several animals in some of the dose groups. However, these findings were considered of dubious significance.

Only a few statistically significant differences were found in the hematology data. The hematocrits in the 12.5 mg/kg dose group were significantly greater than in the vehicle control group. Since the hematocrits in the other dose groups were not significantly higher, this difference does not appear to be compound-related. When compared to the vehicle control group the mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration values were significantly lower in the 50 mg/kg and 100 mg/kg dose groups, respectively. The lack of any other significant changes in the other hematological measurements makes these findings difficult to explain.

CONCLUSIONS

Although MCP caused a few deaths, no definitive clinical chemical, hematological or histological alterations were found. This suggests that death could be due to a transient toxic response associated with cholinesterase inhibition.

RECOMMENDATIONS

Metabolic and pharmacokinetic studies correlating dose and cholinesterase inhibition would aid in the interpretation of data and in the design of dosage regimens for future studies.

REFERENCES

1. Lieske CN, Clark JH, Meyer HG, Lowe JR, Lawson MA, Lennox WJ. The concept and chemistry of organophosphinates as prophylactic agents in organophosphate intoxication. Toxicology Research Projects Directory 1981: 7.

2. Lieske CN, Meyer HG, Clark JH, Lowe JR, King JW. Spontaneous reactivation of bovine erythrocyte acetylcholinesterase inhibited by five organophosphinates. Aberdeen Proving Ground, MD: U.S. Army Medical Bioengineering Research and Development Laboratory, 1979.

3. Horton GL, Lieske CN, Lowe JR. Phosphinate inhibition studies of cholinesterases. Pestic Sci 1978; 9:135-138.

4. Hodgson E, Guthrie FE, eds. Introduction to biochemical toxicity. New York: Elsevier Science Publishing Co., Inc., 1980: 193-223.

5. Dixon WJ, ed. BMDP statistical software. Berkeley: University of California Press, 1981:555-573.

6. White CW, Rodriguez J, Kellner, TP. Acute oral toxicity (LD_{50}) of 4-nitrophenyl monochloromethyl (phenyl) phosphinate (TA009) in male rats. Toxicology Series No. 55. Presidio of San Francisco, CA: Letterman Army Institute of Research. Institute Report No. 191. October 1984.

7. Zimmerman HJ, Henry JB. Serum enzyme determinations as an aid to diagnosis. In: Davidsohn I, Henry JB, eds. Todd-Sanford clinical diagnosis by laboratory methods. Philadelphia: W.B. Saunders Co., 1974:837-869.

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APPENDICES

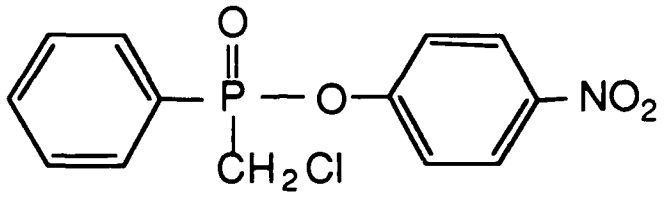
CHEMICAL DATA

Chemical name: 4-Nitrophenyl monochloromethyl (phenyl) phosphinate (MCP)

Lot Number: L-90

LAIR Code: TA009

Structural Formula:



Molecular Formula: $C_{13}H_{11}ClNO_4P$

Physical State: White crystalline solid

Melting Point: 77-78.5° C

Source: Dr. Clair Lieske
US Army Medical Institute of Chemical Defense
Aberdeen Proving Ground, MD 21005

Analytical Data:

Stability: The dosing solutions were assayed for intact and hydrolyzed phosphinate immediately after preparation and dosing. p-Nitrophenol, a product of phosphinate hydrolysis, was quantitated spectrophotometrically at 400 nm using a value of 18,300 for the molar extinction coefficient. Absorbance was measured in accordance with LAIR SOP-OP-STX-49, "Spectrophotometric measurement of p-nitrophenol for phosphinate determination". The concentration of unhydrolyzed phosphinate in the dosing solution was determined from the difference in p-nitrophenol concentraton before and after NaOH hydrolysis. The initial hydrolyzed phosphinate was divided by the total hydrolyzed phosphinate to obtain the percent hydrolysis for each solution. The percent hydrolysis before and after dosing is shown in Table 1.

Concentration: The same analysis described under stability provided information regarding the concentration of the dosing solutions. These results are summarized in Table 2.

TABLE . .e hydrolysis of MCP in the dosing vehicle.

Date	Percent Hydrolysis Before	After	Average
1 Dec 82	7.20	7.36	7.28
2 Dec 82	6.86	6.50	6.68
3 Dec 82	6.86	7.40	7.13
4 Dec 82	5.59	5.78	5.69
5 Dec 82	6.63	7.05	6.84
6 Dec 82	5.88	6.20	6.04
7 Dec 82	6.46	7.13	6.80
8 Dec 82	7.42	7.36	7.39
9 Dec 82	6.42	6.77	6.60
10 Dec 82	5.32	7.00	6.16
11 Dec 82	6.08	6.68	6.38
12 Dec 82	5.94	6.38	6.16
13 Dec 82	5.98	6.89	6.44
14 Dec 82	6.17	6.88	6.53
15 Dec 82	6.24	6.79	6.52

TABLE 2. Actual concentration of MCP in dosing solutions.

Date	Intact MCP (mg/ml) Before	After	Average	% Target
1 Dec 82	13.2	12.0	12.6	90
2 Dec 82	12.3	11.9	12.1	86
3 Dec 82	13.0	12.5	12.8	91
4 Dec 82	12.0	12.4	12.2	87
5 Dec 82	12.5	13.0	12.8	91
6 Dec 82	13.9	11.8	12.9	92
7 Dec 82	12.3	13.5	12.9	92
8 Dec 82	12.6	12.0	12.3	88
9 Dec 82	12.8	13.5	13.2	94
10 Dec 82	11.6	11.5	11.6	83
11 Dec 82	13.1	12.1	12.6	90
12 Dec 82	12.8	12.3	12.6	90
13 Dec 82	12.4	12.5	12.5	89
14 Dec 82	11.5	11.5	11.5	82
15 Dec 82	14.2	14.2	14.2	101

APPENDIX A .luded)

LIST OF TABLES

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TABLE 1

Dosing Scheme and Related Deaths by Group
for
14-Day Subchronic Toxicity of MCP*

Concentration (mg/kg/day)	Group No.	Deaths/ Group Totals
Cage control (0)	1	0/10
Vehicle control (0)	2	0/10
12.5	3	0/9†
25	4	0/10
50	5	1/10
100	6	1/9†

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†one animal misdosed.

TABLE 2

Mean Body Weights

14-Day Subchronic Toxicity of MCP*

	Study Day									
	Q0†	Q2	Q5	Q9	Q12	1	5	9	12	14/15‡
Cage Controls (n=10)	158 ± 1"	178 ± 6	210 ± 4	242 ± 5	267 ± 4	294 ± 4	324 ± 4	351 ± 5	360 ± 5	345 ± 6
Vehicle Controls (n=10)	162 ± 3	175 ± 6	208 ± 4	233 ± 8	249 ± 11	282 ± 8	307 ± 7	329 ± 8¶	334 ± 10¶	317 ± 9¶
12.5 mg/kg (n=9)	160 ± 4	182 ± 4	209 ± 4	242 ± 5	264 ± 6	283 ± 6	317 ± 8	340 ± 9	342 ± 9	321 ± 9
25 mg/kg (n=10)	159 ± 3	182 ± 3	209 ± 3	240 ± 4	256 ± 8	282 ± 6	307 ± 7	326 ± 9	330 ± 10	314 ± 10
50 mg/kg (n=10)	162 ± 2	183 ± 2	209 ± 3	244 ± 6	263 ± 3	284 ± 4	304 ± 3	319 ± 3#	330 ± 4#	312 ± 4#
100 mg/kg (n=9)	161 ± 3	181 ± 4	210 ± 4	244 ± 4	268 ± 5	290 ± 6	301 ± 9**	321 ± 12**	336 ± 13**	317 ± 13**

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Q=quarantine period

‡Fasted overnight

"Mean ± Standard Error

¶The vehicle control group is significantly lower than the cage control group ($p < .05$) by the Student's t-test.

#n=9

**n=8

APPENDIX B (cont.)

TABLE 3

Effects of MCP*
on
Electrolyte Levels in Serum

	Sodium (mg/dl)	Potassium (mEq/L)	Chloride (mEq/L)	Calcium (mg/dl)	Phosphorus (mg/dl)	Magnesium (mg/dl)
Cage Controls (n=10)	148.4 ± 1.6†	6.38 ± 0.12	95.4 ± 1.3	12.85 ± 0.15	9.2 ± 0.3	2.73 ± 0.05
Vehicle Controls (n=10)	149.0 ± 1.9	6.43 ± 0.15	97.3 ± 1.4	12.69 ± 0.25	8.9 ± 0.3	2.89 ± 0.10
12.5 mg/kg (n=9)	150.2 ± 1.9	6.34 ± 0.23	96.9 ± 0.9	12.73 ± 0.25	9.1 ± 0.3	2.87 ± 0.10
25 mg/kg (n=10)	150.3 ± 2.5	6.63 ± 0.23	97.1 ± 1.9	12.67 ± 0.30‡	8.6 ± 0.3‡	2.67 ± 0.09
50 mg/kg (n=9)	148.5 ± 1.9	6.56 ± 0.18	98.2 ± 1.7	12.70 ± 0.20	9.0 ± 0.3	2.73 ± 0.08
100 mg/kg (n=8)	152.0 ± 1.6	6.53 ± 0.12	100.0 ± 1.2	12.32 ± 0.26	8.8 ± 0.4	2.71 ± 0.06

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Mean ± Standard Error

‡n=9

TABLE 4

The Effect of MCP* on Biochemical Constituents of Serum

	Triglycerides (mg/dl)	Cholesterol (mg/dl)	Glucose (mg/dl)	Creatinine (mg/dl)	Blood Urea Nitrogen (mg/dl)	Uric Acid (mg/dl)	Albumin (gm/dl)	Globulin (gm/dl)	Total Protein (gm/dl)	Total Bilirubin (mg/dl)	Serum Iron (ug/dl)
Cage Controls (n=10)	77.3 ± 13.2†	104 ± 6	201 ± 13	0.74 ± 0.03	16.20 ± 0.72	3.1 ± 0.2	5.28 ± 0.09	1.72 ± 0.05	7.00 ± 0.10	0.01 ± 0.02‡	188 ± 23
Vehicle Controls (n=10)	56.2 ± 6.0	92 ± 2	208 ± 13	0.68 ± 0.03	16.79 ± 0.83	3.0 ± 0.2	5.29 ± 0.10	1.68 ± 0.06	6.97 ± 0.12	0.01 ± 0.01	238 ± 36
12.5 mg/kg (n=9)	53.5 ± 7.0	92 ± 5	218 ± 20	0.68 ± 0.03	17.89 ± 0.48	3.0 ± 0.3	5.16 ± 0.17	1.85 ± 0.15	7.01 ± 0.15	0.00 ± 0.01	199 ± 36
25 mg/kg (n=10)	56.4 ± 6.0	95 ± 4	225 ± 13	0.66 ± 0.02	15.05 ± 0.64	3.4 ± 0.2	5.01 ± 0.21	1.91 ± 0.13	6.91 ± 0.16	0.02 ± 0.01	152 ± 24
50 mg/kg (n=9)	59.4 ± 8.7	89 ± 4	223 ± 18	0.69 ± 0.03	15.72 ± 0.42	3.1 ± 0.3	5.17 ± 0.11	1.66 ± 0.06	6.83 ± 0.14	0.01 ± 0.01	143 ± 20
100 mg/kg (n=8)	73.6 ± 16.9	102 ± 5	198 ± 17	0.65 ± 0.02	15.81 ± 0.52	2.8 ± 0.1	4.75 ± 0.19	1.70 ± 0.06	6.45 ± 0.23	0.00 ± 0.00	158 ± 37

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Mean ± Standard Error

‡Median ± Standard Error

APPENDIX B (cont.)

APPENDIX B (cont.)

TABLE 5

Effects of MCP* on Serum Enzyme Activity

	Aspartate Amino-Transferase (I.U.)	Alanine Amino-Transferase (I.U.)	Lactate Dehydrogenase (I.U.)	Creatine Phosphokinase (I.U.)	Alkaline Phospokinase (I.U.)
Cage Controls (n=10)	53.13 ± 1.75†	28.41 ± 1.09	63.13 ± 5.88	130.13 ± 14.07	171.75 ± 9.24
Venicle Controls (n=10)	52.81 ± 1.52	26.98 ± 0.72	75.75 ± 9.52	100.98 ± 10.78	156.78 ± 5.61
12.5 mg/kg (n=9)	57.84 ± 2.65	27.42 ± 1.61	62.38 ± 7.39	112.68 ± 15.14	169.96 ± 7.19
25 mg/kg (n=10)	54.91 ± 1.68	28.72 ± 0.98	70.80 ± 8.22	102.62 ± 6.43	163.62 ± 9.25
50 mg/kg (n=9)	53.65 ± 2.48	26.72 ± 1.57	70.51 ± 8.63	138.23 ± 24.33	143.45 ± 6.78
100 mg/kg (n=8)	55.45 ± 2.96‡	26.73 ± 1.55	58.57 ± 13.62	76.71 ± 6.86	127.93 ± 7.39"

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Mean ± Standard Error

‡n=7

"The 100 mg/kg dose group is significantly lower than the vehicle control group ($p < .05$) by the Dunnett's test.

TABLE 6

Effect of MCP* on Cholinesterase Activity in Plasma, Red Blood Cell and Brain

	Acetylcholinesterase Activity			Butyrylcholinesterase Activity		
	Plasma (I.U.)	Red Blood Cell (I.U.)	Brain (I.U.)	Plasma (I.U.)	Red Blood Cell (I.U.)	Brain (I.U.)
Cage Controls (n=10)	0.38 ± 0.02†	1.74 ± 0.08	7.05 ± 0.49	0.077 ± 0.006	0.630 ± 0.020	0.446 ± 0.018
Vehicle Controls (n=10)	0.37 ± 0.02	1.90 ± 0.06	7.51 ± 0.46	0.071 ± 0.004	0.635 ± 0.018	0.470 ± 0.020
12.5 mg/kg (n=9)	0.41 ± 0.02	1.79 ± 0.03	6.23 ± 0.74	0.073 ± 0.004	0.613 ± 0.020	0.440 ± 0.025
25 mg/kg (n=10)	0.38 ± 0.01	1.86 ± 0.08	6.47 ± 0.26	0.066 ± 0.003	0.568 ± 0.024	0.468 ± 0.024
50 mg/kg (n=9)	0.42 ± 0.02	1.89 ± 0.07	8.03 ± 0.46	0.075 ± 0.003	0.572 ± 0.025	0.437 ± 0.024
100 mg/kg (n=8)	0.39 ± 0.04	1.84 ± 0.06	7.93 ± 0.28	0.073 ± 0.006	0.551 ± 0.022	0.474 ± 0.016

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Mean ± Standard Error

APPENDIX B (cont.)

TABLE 7

Effect of MCP* on Hematological Parameters

	Red Blood Cells	Hemoglobin	Hematocrit	Mean Cell Volume	Mean Corpuscular Hemoglobin	Mean Corp. Hemoglobin Concentra.	Reticulocytes	Platelets	White Blood Cells	White Blood Cell Differential			
										Neutrophils	Lymphocytes	Eosinophils	Monocytes
	($x10^6$/ul)	(g/dl)	(%)	(u^3)	(uug)	(%)	(%)	($x10^6$/ul)	($x10^3$/ul)	($x10^3$/ul)	($x10^3$/ul)	($x10^3$/ul)	($x10^3$/ul)
Cage Controls (n=10)	7.80 ± 0.16†	16.9 ± 0.2	43.8 ± 0.7	59 ± 1	21.7 ± 0.5	38.7 ± 0.8	2.8 ± 0.2	878 ± 52	7.8 ± 0.5	1.1 ± 0.2	6.6 ± 0.4	0.0 ± 0.0	0.1 ± 0.0
Vehicle Controls (n=10)	7.62 ± 0.10	16.6 ± 0.2	41.9 ± 0.7	58 ± 1	21.8 ± 0.3	39.7 ± 0.7	2.9 ± 0.2	855 ± 40	7.7 ± 0.4	1.0 ± 0.1	6.6 ± 0.4	0.0 ± 0.0	0.0 ± 0.0
12.5 mg/kg (n=9)	8.10 ± 0.19	17.0 ± 0.3	45.3 ± 0.9‡	59 ± 0	21.1 ± 0.4	37.7 ± 0.6	3.2 ± 0.2	902 ± 59	7.4 ± 0.3	1.0 ± 0.2	6.2 ± 0.2	0.0 ± 0.0	0.1 ± 0.0
25 mg/kg (n=10)	7.80 ± 0.19	16.9 ± 0.3	42.4 ± 1.0	57 ± 1	21.6 ± 0.2	39.9 ± 0.8	3.3 ± 0.3	836 ± 53	7.3 ± 0.6	0.7 ± 0.1	6.5 ± 0.6	0.0 ± 0.0	0.0 ± 0.0
50 mg/kg (n=9)	8.05 ± 0.16	16.5 ± 0.3	43.4 ± 0.7	56 ± 1	20.5 ± 0.3-	38.0 ± 0.8	3.5 ± 0.2	862 ± 47	7.3 ± 0.4	1.0 ± 0.1	6.2 ± 0.4	0.1 ± 0.0	0.0 ± 0.0
100 mg/kg (n=8)	7.72 ± 0.16	16.2 ± 0.4	43.3 ± 0.8	59 ± 0	21.0 ± 0.4	37.4 ± 0.6¶	3.8 ± 0.2	865 ± 69	7.3 ± 0.3	1.1 ± 0.2	6.1 ± 0.3	0.0 ± 0.0	0.0 ± 0.0

*MCP=4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

†Mean ± Standard Error

‡The 12.5 mg/kg dose group is significantly higher than the vehicle control group ($p < .05$) by the Dunnett's test.

-The 50 mg/kg dose group is significantly lower than the vehicle control group ($p < .05$) by the Dunnett's test.

¶The 100 mg/kg dose group is significantly lower than the vehicle control group ($p < .05$) by the Dunnett's test.

HISTORICAL LISTING OF STUDY EVENTS

Date	Events
17 Nov 82	Animals arrived at LAIR. They were observed for illness, eartagged, weighed, and caged in GLP Suite. Two animals were submitted to the LAIR Pathology Group for quality control necropsy.
18-30 Nov 82	Animals were checked daily.
19,22,26, 29 Nov 82	All animals weighed.
30 Nov 82	Animals removed from quarantine status and dosage level calculated for Groups 2(a) - 6(a).
1 Dec 82	Groups 2(a) - 6(a) dosed. Observations conducted at 1000 hours throughout the study period. Dosage for groups 2(b) - 6(b) calculated.
2 Dec 82	Groups 2-6 (a + b) weighed, dosed and observed. Group 1 weighed and observed. Dose levels calculated for Groups 2-6.
3-5 Dec 82	Groups 2-6 dosed and observed. Group 1 observed.
6 Dec 82	Groups 2-6 weighed, dosed and observed. Group 1 weighed and observed. Dose levels calculated for Groups 2-6.
7 Dec 82	Groups 2-6 dosed with newly calculated dose level and observed. Group 1 observed.
8-9 Dec 82	Groups 2-6 dosed and observed. Group 1 observed.
10 Dec 82	All animals weighed. Dose levels for Groups 2 - 6 recalculated.
10-12 Dec 82	Groups 2-6 dosed with newly calculated dose level and observed. Group 1 observed.
13 Dec 82	Groups 2-6 dosed and observed. Group 1 observed. All animals weighed.
14 Dec 82	Groups 2-6 dosed and observed. Group 1 observed. Food removed from Groups 1(a) - 6(a) at 1630 hours. Twelve animals transferred to metabolic cages.
15 Dec 82	Groups 1(a) - 6(a) observed and weighed at 0730. necropsy Groups 1(a) - 6(a). Blood and tissue samples taken for the measurements specified. Groups 2(b) - 6(b) weighed, dosed, and observed. Group 1b observed. Food removed from Groups 1(b) - 6(b) at 1630 hours.

Date	Events
16 Dec 82	Animals observed and weighed at 0730 hours. Groups 1(b) - 6(b) submitted for necropsy. Blood and tissue samples taken for the measurements specified.

PROCEDURES FOR ANALYTICAL CHEMISTRY

The following are LAIR GLP SOPs for the Analytical Chemistry performed for the study.

1. Calcium - OP-ACH-17
2. Sodium and Potassium - OP-ACH-19
3. Chloride - OP-ACH-20
4. Magnesium - OP-ACH-50
5. Phosphorus - OP-ACH-18
6. Glucose - OP-ACH-7
7. Cholesterol - OP-ACH-11
8. Triglycerides - OP-ACH-9
9. Creatinine - OP-ACH-15
10. Blood Urea Nitrogen - OP-ACH-16
11. Uric Acid - OP-ACH-14
12. Albumin - OP-ACH-12
13. Total Protein - OP-ACH-13
14. Total Bilirubin - OP-ACH-8
15. Serum Iron - OP-ACH-22
16. Aspartate Amino-Transferase - OP-ACH-4
17. Alanine Amino-Transferase - OP-ACH-3
18. Lactate Dehydrogenase - OP-ACH-5
19. Creatine Phosphokinase - OP-ACH-6
20. Alkaline Phosphatase - OP-ACH-10
21. Acetyl Cholinesterase - OP-ACH-30 and OP-ACH-46
22. Butyryl Cholinesterase - OP-ACH-52

Globulin values were calculated by subtracting the albumin values from the total protein values.

DEVIATIONS FROM THE ORIGINAL PROTOCOL

1. On 10 Dec 82 dose volumes were recalculated based on the weights taken that day. Normally, the new volumes were not used until the following day. However, this time the new volumes were given the same day they were calculated.

2. On 3 Dec 82 the cage control animals were overlooked when observations were performed.

3. According to the original protocol the vehicle was to be 21.5% Tween 80™, 18.5% ethanol, 37.5% 50mM citrate buffer (pH 3.2), and 22.5% water. The test compound was more susceptible to hydrolysis than previous phosphinate compounds tested, so the vehicle was changed to 20% Tween 80™, 10% ethanol, and 70% 50mM citrate buffer (pH 3.0) which increased the stability of the test compound.

Coding for Clinical Signs

-	Normal
*	Observation not performed
A	Aggressive
B	Brown Urine
C	Rough Coat
D	Diarrhea
E	Excited
F	Decreased Respiratory Rate
G	Increased Respiratory Rate
H	Hunched Posture
I	Inactive or Sluggish
J	Decreased Respiratory Depth
K	Increased Respiratory Depth
L	Loss of Equilibrium
M	Clear Stain Perianal
N	Toe Nail Bleeding
O	Orange Stain Perianal
P	Piloerection
Q	Irritable
R	Red Stain/Material Head/Neck
S	Yellow/Clear Stain/Material Mouth/Front Legs or Salivation
T	Hair Loss
U	Scab
W	Sound Production
X	Dead
Y	Yellow Stain/Material Perianal/Ventral

INDIVIDUAL CLINICAL SIGNS

Group	Animal ID	Days of Study														
		0	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Group 1 - Cage Control	82D00986	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-
	82D00992	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-
	82D00993	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-
	82D01000	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-
	82D01007	-	-	*	-	-	-	-	-	-	-	-	-	-	-	-
	82D01010	-	*	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01022	-	*	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01025	-	*	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01035	-	*	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01041	-	*	-	-	-	-	-	-	-	-	-	-	-	-	-
Group 2 - Vehicle Control	82D00977	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D00980	E	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D00989	-	-	-	-	N	-	-	-	-	-	U	U	U	U	U
	82D00994	-	-	-	-	U	U	U	U	U	U	U	U	U	U	U
	82D00997	-	-	-	-	-	-	-	-	-	-	Q	-	-	-	-
	82D01001	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01008	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01011	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01026	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01028	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

INDIVIDUAL CLINICAL SIGNS

Group	Animal ID	Days of Study														
		0	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Group 3 - 12.5 mg/kg	82D00981	-	-	-	-	-	-	-	-	-	-	-	T	U	TU	TU
	82D00983	-	-	-	-	-	-	-	-	-	FI	-	-	-	-	-
	82D00985	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01004	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01012	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01018	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01021	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01032	E	-	S	-	-	-	-	-	-	-	-	-	-	-	-
	82D01039	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Group 4 - 25 mg/kg	82D00974	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D00996	-	S	-	-	-	-	-	-	-	-	-	I	-	-	R
	82D00998	-	E	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D00999	-	-	-	-	-	-	-	-	-	I	-	ID	ID	ID	-
	82D01005	R	-	-	-	-	-	-	-	-	I	-	I	-	-	-
	82D01014	Q	-	Q	Q	Q	Q	Q	-	Q	Q	QLI	Q	Q	-	-
	82D01023	-	-	-	-	-	-	-	-	I	-	IL	I	-	I	I
	82D01027	IF	-	-	-	-	-	-	-	-	-	F	FI	I	I	-
	82D01033	-	-	-	-	-	-	L	-	SY	SY	SY	SY	SY	SY	S
	82D01037	GJE	-	-	-	-	-	-	-	-	-	-	-	-	-	-

APPENDIX F-1 (cont.)

APPENDIX F-1 (concluded)

INDIVIDUAL CLINICAL SIGNS

Group	Animal ID	Days of Study														
		0	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Group 5 - 50 mg/kg	82D00976	-	-	-	I	-	-	-	-	-	-	-	-	I	I	I
	82D00984	-	-	-	-	-	-	-	X							
	82D00991	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
	82D01003	-	-	-	-	-	-	-	-	-	-	-	S	S	SL	-
	82D01013	-	-	-	-	-	-	-	-	-	-	-	E	I	-	-
	82D01020	-	-	-	-	-	-	-	-	-	-	SE	SE	SE	S	I
	82D01029	E	-	-	-	-	-	SYI	-	E	-	-	SI	-	SIRPH	-
	82D01034	E	-	-	-	-	-	-	E	-	-	-	-	-	ES	-
	82D01036	-	-	-	-	-	-	-	-	Y	Y	YS	Y	YSPW CEK	YPW	S
	82D01038	-	-	-	-	-	-	-	-	S	-	-	I	SI	IM	-
Group 6 - 100 mg/kg	82D00975	BY	Y	I	OR	ISY	ISY	SY	SY	R	R	RS	RS	R	YI	-
	82D00979	-	G	I	I	-	-	I	I	Y	I	I	IY	IYS	IYS	Y
	82D00987	S	S	I	-	E	-	-	-	-	S	I	I	I	I	-
	82D00988	-	U	-	ISL FHP	IR	IFHP	I	IH	T	AY	IR	RTY	RTY	LPRTY	LY
	82D00990	-	-	X												
	82D01015	-	IP	IP	I	IP	I	I	-	IS	-	I	-	-	M	-
	82D01017	-	-	-	-	-	-	I	E	ISQ	Q	Q	Q	EQP	QPY	-
	82D01030	-	IP	IPY	I	IPYF	-	IPYS	Y	IYS	IYS	YSW	SW	PYSW	YFR	-
	82D01040	Y	I	-	-	-	-	I	-	-	-	-	-	-	IR	-

GLP Study #82034

14-Day Subchronic Oral Toxicity in Male Rats
of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

INDIVIDUAL BODY WEIGHTS

Group	Animal ID	QDay0	QDay2	QDay5	QDay9	QDay12	Day1	Day5	Day9	Day12	SacDay
1	82D00986	157	183	209	242	261	291	307	335	336	318
	82D00992	156	180	206	242	265	295	312	338	351	336
	82D00993	158	181	204	241	260	283	317	342	350	335
	82D01000	159	180	211	247	266	293	327	361	367	351
	82D01007	151	129	191	200	255	278	314	341	346	331
	82D01010	159	188	220	256	277	302	334	362	370	354
	82D01022	167	197	233	264	286	308	340	364	375	362
	82D01025	160	182	216	251	282	309	344	361	394	380
	82D01035	157	184	214	245	269	299	330	353	361	352
	82D01041	153	173	198	232	253	279	311	333	345	331
2	82D00977	177	156	218	252	262	291	330	357	367	349
	82D00980	156	138	179	160	151	216	260	287	295	283
	82D00989	147	170	197	225	238	264	280	280	283	247
	82D00994	166	169	209	242	262	289	305	334	343	316
	82D00997	161	186	215	247	267	301	326	344	347	328
	82D01001	157	199	205	233	258	286	315	334	342	325
	82D01006	157	183	216	248	266	300	326	337	346	332
	82D01011	174	198	220	247	271	303	316	351	355	342
	82D01026	174	155	211	239	257	282	311	329	338	319
	82D01028	149	175	205	236	262	286	305	336	342	325
3	82D00981	171	196	220	246	270	291	306	325	332	310
	82D00983	167	191	211	248	270	293	326	361	360	337
	82D00985	151	175	199	230	245	266	298	317	318	295
	82D01004	153	177	206	245	267	279	320	342	347	325
	82D01012	175	199	230	266	289	291	341	359	354	335
	82D01018	156	176	209	250	280	306	348	364	388	369
	82D01021	170	187	215	244	267	292	325	349	352	331
	82D01032	145	162	189	213	233	246	269	288	287	272
	82D01039	151	177	201	235	259	282	314	339	339	323

APPENDIX F-2 (concluded)

GLP Study #82034 — 14-Day Subchronic Oral Toxicity in Male Rats of 4-Nitrophenyl Monochlo-omethyl (Phenyl) Phosphinate — PAGE

INDIVIDUAL BODY WEIGHTS

Group	Animal ID	QDay0	QDay2	QDay5	QDay9	QDay12	Day1	Day5	Day9	Day12	SacDay
4	82D00974	155	170	199	227	245	264	273	284	286	269
	82D00996	164	198	225	260	265	302	329	356	361	347
	82D00998	160	182	208	236	190	243	269	279	275	258
	82D00999	165	193	218	256	277	301	339	364	378	358
	82D01005	139	166	195	229	255	279	300	335	343	323
	82D01014	159	180	205	234	255	277	302	315	318	305
	82D01023	172	192	215	244	268	293	316	330	339	321
	82D01027	160	185	213	239	264	285	308	329	334	322
	82D01033	165	183	211	248	269	297	323	343	334	320
	82D01037	153	175	201	231	256	277	307	325	334	320
5	82D00976	152	170	195	230	255	270	299	324	330	319
	82D00984	159	186	213	242	259	277	298	---	---	---
	82D00991	165	184	206	269	263	283	307	326	346	326
	82D01003	168	192	218	253	272	296	314	326	324	306
	82D01013	165	178	202	229	246	268	290	306	321	295
	82D01020	165	180	209	244	265	284	309	317	335	311
	82D01029	166	190	216	247	274	300	310	311	329	320
	82D01034	167	193	221	245	274	294	308	324	341	329
	82D01036	165	182	211	243	273	295	312	328	327	301
	82D01038	152	174	197	221	247	271	292	308	310	296
6	82D00975	172	188	216	237	263	274	263	276	292	271
	82D00979	152	158	197	231	249	275	300	323	326	305
	82D00987	154	175	198	235	254	271	286	299	310	290
	82D00988	157	180	207	248	276	297	297	323	353	323
	82D00990	156	184	210	242	264	276	---	---	---	---
	82D01015	166	186	219	258	284	312	331	360	376	361
	82D01017	173	193	230	267	298	324	348	375	392	376
	82D01030	158	175	201	233	257	290	286	286	301	281
	82D01040	165	188	212	242	267	291	300	327	342	326

INDIVIDUAL SERUM ELECTROLYTE LEVELS

Group	Animal ID	Sodium	Potassium	Chloride	Calcium	Phosphorous	Magnesium
		mg/dl	mEq/L	mEq/L	mg/dl	mg/dl	mg/dl
1	82D00986	147.6	5.89	98.3	12.33	8.9	2.62
	82D00992	154.3	6.72	100.5	11.92	9.5	2.58
	82D00993	154.4	6.53	99.0	12.95	9.0	2.62
	82D01000	154.8	7.08	97.6	12.53	10.1	2.66
	82D01007	142.6	6.29	95.5	12.94	9.3	2.81
	82D01010	150.4	6.49	97.9	12.99	8.4	2.99
	82D01022	147.4	6.10	94.4	13.01	7.9	2.69
	82D01025	141.4	6.09	88.5	13.65	8.3	2.75
	82D01035	143.1	5.94	89.5	13.15	9.6	2.63
	82D01041	148.1	6.70	92.5	13.07	10.5	2.96
2	82D00977	149.8	6.67	100.4	12.54	9.7	2.71
	82D00980	151.9	6.49	99.8	11.98	10.5	3.07
	82D00989	157.2	6.72	103.4	12.39	8.7	2.58
	82D00994	157.5	6.39	101.1	12.92	10.0	2.53
	82D00997	145.2	6.39	98.3	12.48	9.3	2.65
	82D01001	153.1	6.87	98.1	11.23	8.5	3.06
	82D01008	142.8	6.92	92.7	12.98	8.0	3.53
	82D01011	148.0	5.21	96.5	12.77	8.1	2.88
	82D01026	144.0	6.40	93.0	13.59	8.4	2.67
	82D01028	140.4	6.19	89.9	14.04	7.9	3.23
3	82D00981	147.4	6.07	97.6	12.16	9.9	2.84
	82D00983	150.6	7.12	100.5	11.96	9.9	2.83
	82D00985	150.0	5.86	98.3	12.25	10.0	2.52
	82D01004	151.0	6.18	94.7	14.41	9.0	2.98
	82D01012	146.8	6.01	93.7	12.32	8.4	2.95
	82D01018	150.5	5.84	96.9	13.34	7.7	3.20
	82D01021	163.8	7.86	101.1	12.81	9.2	3.30
	82D01032	145.6	5.99	95.4	12.52	8.4	2.40
	82D01039	145.7	6.14	93.8	12.84	9.6	2.62

GLP Study #82034

14-Day Subchronic Oral Toxicity in Male Rats
of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

INDIVIDUAL SERUM ELECTROLYTE LEVELS

Group	Animal ID	Sodium	Potassium	Chloride	Calcium	Phosphorous	Magnesium
		mg/dl	mEq/L	mEq/L	mg/dl	mg/dl	mg/dl
4	82D00974	158.7	6.65	103.6	-----	8.4	2.85
	82D00996	145.9	7.21	95.0	12.61	9.6	2.88
	82D00998	164.7	7.82	107.5	12.35	6.8	2.62
	82D00999	150.3	7.43	95.5	12.83	9.8	2.23
	82D01005	158.4	6.69	104.8	10.57	9.6	2.40
	82D01014	146.4	6.02	94.1	12.68	7.5	3.18
	82D01023	146.9	5.35	91.8	12.91	7.2	2.51
	82D01027	147.3	6.63	96.0	13.37	8.1	2.54
	82D01033	144.3	6.57	93.7	13.09	---	2.67
	82D01037	139.7	5.97	88.5	13.66	8.6	2.83
5	82D00976	157.2	7.09	104.4	11.95	8.5	2.74
	82D00991	148.1	5.92	97.9	12.40	11.1	2.46
	82D01003	153.9	7.36	103.2	12.50	9.9	2.79
	82D01013	156.1	7.31	102.0	12.19	8.5	2.76
	82D01020	144.8	6.37	94.5	14.01	8.0	3.17
	82D01029	147.6	6.29	102.8	12.85	8.4	2.34
	82D01034	144.6	6.11	95.7	12.43	8.5	2.63
	82D01036	142.3	6.09	91.8	13.16	8.8	2.69
	82D01038	142.3	6.48	91.5	12.82	9.2	3.00
6	82D00975	156.8	7.05	104.1	11.65	9.0	2.84
	82D00979	154.4	6.73	102.4	11.57	7.7	2.69
	82D00987	157.8	6.63	102.3	12.39	8.5	2.69
	82D00988	156.0	6.70	102.0	12.05	11.5	2.61
	82D01015	148.1	6.25	96.2	13.61	8.2	2.99
	82D01017	147.2	6.11	96.4	13.13	8.1	2.71
	82D01030	147.5	6.63	100.7	11.74	8.7	2.28
	82D01040	148.5	6.16	95.5	12.44	8.8	2.90

INDIVIDUAL SERUM BIOCHEMICAL LEVELS

Group	Animal ID	Triglycerides	Cholesterol	Glucose	Creatinine	Blood Urea Nitrogen	Uric Acid	Albumin	Globulin	Total Protein	Total Bilirubin	Serum Iron
		mg/dl	mg/dl	mg/dl	mg/dl	mg/dl	mg/dl	gm/dl	gm/dl	gm/dl	mg/dl	ug/dl
1	82D00986	175	109	175	0.68	18.57	1.7	4.86	1.99	6.85	0.00	146
	82D00992	131	74	131	0.71	13.38	2.4	5.15	1.48	6.63	0.00	310
	82D00993	171	89	171	0.61	13.73	3.2	5.11	1.57	6.68	0.00	116
	82D01000	206	95	206	0.77	18.51	2.9	5.00	1.74	6.74	0.00	256
	82D01007	155	122	155	0.67	15.49	4.1	5.05	1.70	6.75	0.08	226
	82D01010	230	123	230	0.80	17.00	3.7	5.56	1.93	7.49	----	278
	82D01022	219	99	219	0.69	14.36	3.0	5.42	1.88	7.30	0.01	114
	82D01025	236	100	236	0.72	13.85	2.9	5.70	1.71	7.41	0.10	112
	82D01035	270	130	270	0.83	17.95	2.9	5.35	1.65	7.00	0.05	164
	82D01041	216	94	216	0.87	19.16	3.7	5.56	1.58	7.14	0.03	160
2	82D00977	179	96	179	0.65	13.60	2.6	5.20	1.64	6.84	0.00	430
	82D00980	178	93	178	0.62	14.71	3.1	5.01	1.52	6.53	0.01	188
	82D00989	207	83	207	0.67	20.44	2.9	4.98	2.06	7.04	0.00	110
	82D00994	184	88	184	0.65	16.15	2.6	5.63	1.50	7.13	0.00	272
	82D00997	180	90	180	0.53	17.06	2.6	5.21	1.78	6.99	0.00	82
	82D01001	206	87	206	0.80	15.27	3.0	4.82	1.44	6.26	0.04	220
	82D01008	248	100	248	0.85	14.50	4.6	5.49	1.95	7.44	0.03	156
	82D01011	171	86	171	0.72	17.86	2.1	5.22	1.63	6.85	0.01	208
	82D01026	228	88	228	0.60	16.52	2.3	5.56	1.60	7.16	0.04	376
	82D01028	298	103	298	0.67	21.75	4.0	5.77	1.67	7.44	0.02	342
3	82D00981	157	93	157	0.64	20.00	1.8	4.82	1.98	6.80	0.00	170
	82D00983	125	93	125	0.64	16.05	2.9	4.80	1.65	6.45	0.00	106
	82D00985	191	83	191	0.53	16.63	2.3	4.94	1.80	6.74	0.00	286
	82D01004	179	92	179	0.67	18.73	3.8	6.23	1.63	7.86	0.01	108
	82D01012	263	83	263	0.80	16.46	3.2	5.11	1.91	7.02	0.00	234
	82D01018	280	126	280	0.73	17.47	3.0	4.44	2.95	7.39	0.01	286
	82D01021	300	77	300	0.70	18.08	4.8	5.49	1.58	7.07	0.05	396
	82D01032	221	78	221	0.63	19.94	2.2	5.21	1.35	6.56	0.06	118
	82D01039	243	97	243	0.78	17.62	3.1	5.42	1.76	7.18	0.00	88

GLP Study #82034

14-Day Subchronic Oral Toxicity in Male Rats
of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

INDIVIDUAL SERUM BIOCHEMICAL LEVELS

Group	Animal ID	Triglycerides	Cholesterol	Glucose	Creatinine	Blood Urea Nitrogen	Uric Acid	Albumin	Globulin	Total Protein	Total Bilirubin	Serum Iron
		mg/dl	mg/dl	mg/dl	mg/dl	mg/dl	mg/dl	gm/dl	gm/dl	gm/dl	mg/dl	ug/dl
4	82D00974	181	79	181	0.63	15.52	2.2	3.70	2.90	6.60	0.00	344
	82D00996	188	113	188	0.63	13.51	4.1	5.03	1.93	6.96	0.04	180
	82D00998	204	78	204	0.61	17.00	3.3	5.12	1.68	6.80	0.19	94
	82D00999	214	108	214	0.61	14.79	3.1	5.43	1.63	7.06	0.03	104
	82D01005	165	90	165	0.58	11.49	3.7	4.26	1.52	5.78	0.00	96
	82D01014	254	104	254	0.80	16.07	3.5	5.39	1.92	7.31	0.00	136
	82D01023	283	91	283	0.65	13.30	3.4	5.55	1.65	7.20	0.04	124
	82D01027	222	89	222	0.72	13.96	3.3	5.58	1.72	7.30	0.03	120
	82D01033	258	89	258	0.65	16.72	3.4	4.48	2.18	6.66	0.01	118
	82D01037	278	108	278	0.74	18.12	3.6	5.52	1.93	7.45	0.00	208
5	82D00976	164	104	164	0.66	15.75	2.9	4.92	1.64	6.56	0.00	284
	82D00991	160	77	160	0.67	14.58	2.5	4.84	1.59	6.43	0.01	90
	82D01003	187	81	187	0.59	14.45	3.5	5.09	1.43	6.52	0.02	98
	82D01013	231	85	231	0.79	16.04	3.3	5.12	1.50	6.62	0.00	148
	82D01020	305	114	305	0.74	17.82	4.6	5.66	1.78	7.44	0.04	178
	82D01029	192	77	192	0.56	15.59	2.2	4.89	1.54	6.43	0.02	124
	82D01034	260	89	260	0.66	13.97	3.2	4.95	1.86	6.81	0.00	120
	82D01036	210	92	210	0.73	17.21	2.3	5.46	1.97	7.43	0.01	102
	82D01038	292	80	292	0.79	16.09	3.7	5.59	1.67	7.26	0.00	142
6	82D00975	123	110	123	0.64	14.83	2.6	4.11	1.71	5.82	0.01	110
	82D00979	183	91	183	0.71	18.75	2.7	4.73	1.69	6.42	0.00	406
	82D00987	186	78	186	0.57	14.60	2.7	4.92	1.84	6.76	0.01	180
	82D00988	153	99	153	0.55	15.23	2.8	4.23	1.74	5.97	0.00	140
	82D01015	256	121	256	0.72	16.59	3.0	5.47	1.66	7.13	0.00	100
	82D01017	240	99	240	0.70	16.24	2.7	5.39	1.86	7.25	0.00	78
	82D01030	182	96	182	0.60	16.08	2.5	4.12	1.31	5.43	0.00	110
	82D01040	258	115	258	0.67	14.13	3.7	5.03	1.77	6.80	0.00	136

INDIVIDUAL ENZYME ACTIVITIES (I.U.)

Group	Animal ID	Aspartate Amino-Transfer.	Alanine Amino-Transfer.	Lactate Dehydro.	Creatine Phospho.	Alkaline Phospha.	Acetylcholinesterase			Butyrylcholinesterase		
							Plasma	RBC	Brain	Plasma	RBC	Brain
1	82D00986	48.10	29.60	53.67	92.63	127.38	0.40	1.83	7.05	0.065	0.662	0.487
	82D00992	56.80	29.26	38.26	120.35	240.94	0.38	1.53	6.97	0.073	0.573	0.337
	82D00993	47.00	35.37	46.70	117.39	172.41	0.46	1.51	5.19	0.102	0.609	0.491
	82D01000	54.90	26.35	78.92	153.62	165.47	0.32	1.67	5.67	0.065	0.543	0.411
	82D01007	51.68	24.47	91.79	101.88	169.42	0.47	1.31	7.78	0.109	0.538	0.451
	82D01010	52.24	25.54	55.43	75.54	158.34	0.34	2.18	7.75	0.065	0.710	0.511
	82D01022	57.11	29.12	53.74	87.04	175.96	0.39	1.90	9.41	0.073	0.650	0.505
	82D01025	55.73	27.62	92.70	182.00	153.04	0.40	1.96	9.34	0.087	0.710	0.464
	82D01035	44.67	24.70	63.44	209.43	165.66	0.31	1.79	5.26	0.058	0.648	0.395
	82D01041	63.07	32.10	56.69	161.46	188.85	0.36	1.71	6.05	0.073	0.659	0.411
2	82D00977	50.90	23.77	114.09	92.60	148.87	0.28	2.01	7.09	0.051	0.582	0.489
	82D00980	55.80	29.26	64.29	64.01	190.16	0.40	1.90	5.59	0.073	0.585	0.506
	82D00989	57.10	25.72	51.35	99.46	150.68	0.39	1.83	6.23	0.065	0.582	0.452
	82D00994	46.40	25.72	31.44	61.69	143.30	0.42	1.61	6.69	0.073	0.585	0.404
	82D00997	51.00	26.07	127.17	105.93	147.99	0.35	1.63	8.55	0.065	0.592	0.364
	82D01001	44.98	24.03	94.32	138.39	162.29	0.36	2.26	10.09	0.080	0.655	0.444
	82D01008	58.67	30.60	90.24	154.39	148.19	0.30	2.07	8.57	0.065	0.670	0.452
	82D01011	57.44	28.27	67.88	120.77	144.02	0.36	1.88	5.79	0.073	0.725	0.599
	82D01026	50.25	28.64	50.08	51.77	187.57	0.35	2.05	8.13	0.073	0.684	0.501
	82D01028	55.56	27.71	66.68	120.80	144.74	0.44	1.79	8.34	0.094	0.690	0.484
3	82D00981	48.00	25.60	40.94	69.39	157.90	0.36	1.65	2.66	0.065	0.494	0.483
	82D00983	59.70	25.05	43.82	63.94	212.87	0.40	1.76	5.31	0.080	0.617	0.399
	82D00985	50.70	24.43	39.32	57.68	171.97	0.48	1.86	9.56	0.073	0.655	0.541
	82D01004	59.10	25.68	77.51	99.00	174.02	0.44	1.74	6.97	0.073	0.552	0.361
	82D01012	52.72	24.64	49.03	173.52	148.13	0.30	1.89	5.17	0.051	0.684	0.469
	82D01018	67.49	36.90	56.20	153.55	182.30	0.38	1.66	8.74	0.080	0.662	0.511
	82D01021	51.61	22.29	90.52	94.39	141.66	0.41	1.89	4.02	0.080	0.648	0.307
	82D01032	71.64	33.71	99.11	138.18	182.24	0.41	1.90	5.96	0.058	0.592	0.432
	82D01039	59.62	28.49	64.99	164.45	158.56	0.50	1.74	7.67	0.094	0.610	0.454

INDIVIDUAL ENZYME ACTIVITIES (I.U.)

Group	Animal ID	Aspartate Amino-Transfer.	Alanine Amino-Transfer.	Lactate Dehydro.	Creatine Phospho.	Alkaline Phospha.	Acetylcholinesterase			Butyrylcholinesterase		
							Plasma	RBC	Brain	Plasma	RBC	Brain
4	82D0[illegible]	51.90	22.36	73.08	129.25	161.40	0.35	1.88	7.19	0.051	0.520	0.485
	82D0[illegible]	52.10	30.21	130.41	75.82	152.22	0.40	1.75	7.07	0.073	0.492	0.554
	82D[illegible]	60.50	32.45	92.35	87.46	213.72	0.40	1.81	5.62	0.073	0.492	0.509
	82D[illegible]9	48.50	27.49	69.28	114.12	172.03	0.37	1.61	7.38	0.058	0.443	0.428
	82D01005	48.45	26.61	80.04	99.25	145.83	0.36	1.30	6.68	0.073	0.573	0.415
	82D01014	56.29	30.51	49.66	120.52	158.36	0.35	2.11	6.48	0.058	0.626	0.494
	82D01023	61.43	26.81	62.04	114.40	129.75	0.47	2.18	4.65	0.065	0.631	0.293
	82D01027	60.94	32.12	61.69	99.42	205.68	0.40	1.98	6.69	0.073	0.655	0.473
	82D01033	50.00	30.87	42.55	118.13	123.33	0.32	2.07	6.73	0.058	0.631	0.542
	82D01037	59.00	27.77	46.92	67.81	173.86	0.37	1.87	6.22	0.073	0.621	0.488
5	82D00976	51.40	24.57	121.19	97.56	184.78	0.41	1.98	9.01	0.080	0.533	0.544
	82D00991	53.50	27.23	81.17	295.10	154.25	0.34	1.74	8.77	0.058	0.479	0.382
	82D01003	48.80	23.58	60.07	80.54	135.18	0.38	1.56	5.76	0.073	0.479	0.338
	82D01013	53.01	20.83	65.20	116.80	146.77	0.40	1.59	7.92	0.065	0.516	0.420
	82D01020	70.59	37.04	62.60	83.28	132.23	0.42	2.15	9.96	0.073	0.655	0.433
	82D01029	58.98	30.07	98.33	225.89	112.66	0.49	2.02	9.27	0.087	0.585	0.507
	82D01034	46.29	26.28	35.59	108.53	137.02	0.46	1.87	7.34	0.073	0.659	0.519
	82D01036	47.09	23.48	52.05	108.14	132.09	0.52	2.15	7.88	0.087	0.584	0.418
	82D01038	53.21	27.40	58.38	128.26	156.09	0.40	1.98	6.32	0.080	0.659	0.371
6	82D00975	57.40	22.26	149.89	115.42	91.03	0.25	1.71	9.28	0.058	0.497	0.488
	82D00979	59.60	28.68	50.50	84.05	125.65	0.40	1.77	8.10	0.073	0.514	0.522
	82D00987	48.90	26.04	43.54	58.77	153.73	0.42	1.93	7.57	0.080	0.594	0.433
	82D00988	-----	21.87	30.60	57.15	118.80	0.40	1.49	7.72	0.087	0.449	0.457
	82D01015	69.10	35.06	41.71	77.05	134.36	0.43	2.03	6.96	0.087	0.573	0.473
	82D01017	44.92	26.15	36.86	86.09	137.79	0.33	2.01	8.72	0.051	0.606	0.555
	82D01030	55.47	24.03	68.02	75.47	111.24	0.29	1.90	7.99	0.051	0.543	0.434
	82D01040	52.77	29.76	47.41	59.65	150.84	0.59	1.90	7.06	0.094	0.631	0.426

Pathology Report

Fourteen Day Sub-chronic Toxicity Study of 4-Nitrophenyl Monochloromethyl(phenyl)phosphinate in Male Albino Sprague-Dawley Rats, Study 82-034

1. Introduction.

The objective of this study was to determine the sub-chronic effects of 4-Nitrophenyl Monochloromethyl(phenyl)phosphinate when administered daily for 14 days (oral gavage) in male Sprague-Dawley rats. Each animal was randomly assigned to one of 6 dose groups of 10 animals each (5 in each subgroup).

Cage controls - groups 1A & 1B
Vehicle* controls - groups 2A & 2B
12.5 mg/kg/day - groups 3A & 3B
25 mg/kg/day - groups 4A & 4B
50 mg/kg/day - groups 5A & 5B
100 mg/kg/day - groups 6A & 6B

After 14 days on test, the rats were submitted for necropsy. Following anesthesia with pentobarbitol sodium, administered by intraperitoneal injection, blood was collected from the right ventricle of each rat and submitted for hematologic examination [red blood cell count (RBC), hemoglobin concentration (Hb), hematocrit (Hct), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), white blood cell count (WBC), WBC differential and blood cell morphology, platelet count, and reticulocyte count]. Additional blood was submitted to Analytical Chemistry Services Group, Division of Research Support, for chemical analyses. All rats were killed by exsanguination and gross necropsy examinations were performed. Portions of anterior cerebrum (unfixed) were submitted to Analytical Chemistry Services Group, Division of Research Support, for cholinesterase determinations. Tissue specimens from major organs and systems were fixed in 10% neutral buffered formalin (except the eyes which were fixed in Karnovsky's solution) for subsequent microscopic examination. Tissues were embedded in paraffin, sectioned at approximately 6 microns thickness and stained with hematoxylin and eosin. All tissues itemized in SOP OP-PSG-12 were examined microscopically in the cage controls, vehicle controls, and the 100 mg/kg dosage level. In the 50 mg/kg, 25 mg/kg, and 12.5 mg/kg dosage levels, only hearts, livers, and kidneys were examined microscopically. In addition, organs with gross lesions were examined microscopically.

*Vehicle: 20% Polysorbate 80 (Tween 80), 10% Ethanol, 70% 50 mM Citrate Buffer.

2. Results, interpretation, and discussion.

The gross and/or microscopic findings are itemized in Incidence Tables 1 - 3.

a. Table 1 tabulates the incidence and severity of lesions observed grossly or microscopically in each rat.

b. Table 2 tabulates group gross necropsy observations.

c. Table 3 tabulates the group histopathologic observations.

Hematology: One way analysis of variance followed by Dunnett's test if applicable, was performed on white cell differentials, MCV's, MCH's and MCHC's, hematocrits, RBC, WBC, reticulocyte, and platelet counts to determine if there were any differences among the vehicle control and each of 12.5, 25, 50, and 100 mg/kg dose groups. The mean hematocrit was significantly greater in the 12.5 mg/kg rats. The mean corpuscular hemoglobin was significantly lower in the 50 mg/kg rats. The mean corpuscular hemoglobin concentration was significantly lower in the 100 mg/kg animals.

d. Gross necropsy:

There were four spontaneous deaths during the course of the study; one of which was a group 3, 12.5 mg/kg rat #33084 on day 10. At necropsy, the presence of oily, reddish-tinged staining around the muzzle of this rat suggested that it had aspirated the test material. A group 5, 50 mg/kg rat #33062 was found dead on day 7. The only gross finding in this animal was a diffusely reddened glandular stomach mucosa. Two group 6, 100 mg/kg rats (#33068 and #33093) died on days 2 and 12 respectively. Rat #33068 when necropsied had a soft brain and a slightly distended mucoid filled small intestine suggesting some degree of autolysis. This rat's stomach's glandular mucosa was also reddened, however. Rat #33093 had several gross necropsy findings suggestive of aspiration (red oily material around the muzzle, firm dark noncollapsed lung lobes) as well as esophageal rupture and intrathoracic installation of test material (dark red subserosal esophageal focus, oily material in the thorax).

Necropsy findings of spontaneously dying rats would therefore suggest that two of the rats died as a direct result of a dosing accident (#33084, #33093) while the other two rats (#33062 and #33068) whose mode of death is speculative may have exhibited signs of mild gastric irritation.

Of the sixty animals in the study; 10 of 10 cage controls, 10 of 10 vehicle controls, 9 of 10 12.5 mg/kg rats, 10 of 10 25 mg/kg rats, 9 of 10 50 mg/kg rats, and 8 of 10 100 mg/kg rats survived to study

termination at which time they were necropsied. Gross necropsy observations were minimal and considered unrelated to compound administration. They consisted (see tables 1 & 2) of: dilated renal pelvises in one vehicle control and one 100 mg/kg rat; thickening of the splenic capsule in one 12.5 mg/kg rat; a focal skin abrasion in another 12.5 mg/kg rat; as well as yellow-brown and red-brown pulmonary foci in one 25 mg/kg and one 50 mg/kg rat respectively.

There were no gross findings in terminally sacrificed rats that might indicate any degree of gastro-intestinal irritation.

e. Microscopic findings:

The majority of histopathologic lesions observed in tissues from animals surviving to terminal sacrifice were considered unrelated to treatment due to frequency of occurance, distribution among dose groups, and incidence rates in normal healthy Sprague Dawley rats.

Peritracheal hemorrhage in one of 6 100 mg/kg tracheas as well as esophagitis and periesophagitis noted in two of seven 100 mg/kg rats were most probably related to the gavage procedure.

Interstitial pneumonitis in 2 of 10 vehicle controls, the one histologically examined 50 mg/kg rat lung and 2 of 8 examined 100 mg/kg rat lungs may well have been related to the gavage procedure with associated aspiration of small quantities of test material and/or concurrent disease.

Hemorrhage and/or erythrophagocytosis was observed in 4 of 8 histologcally examined mesenteric lymph nodes in the 100 mg/kg group.

Portally oriented subacute hepatitis was present in the livers of 2 of 9 and 3 of 8 (50 and 100 mg/kg respectively) histologically examined rats.

There was an increase, although not dose related, in renal tubular mineralization in all four treatment groups.

Rats that died spontaneously had a few of the above-noted lesions. Renal tubular mineralization was present in high dose (100 mg/kg) in rat #33068. Periportal subacute hepatitis was present in group 6 (100 mg/kg) rat #33068 and group 5 (50 mg/kg) rat #33062. Hepatic necrosis was seen in 50 mg/kg rat #33062. Hemorrhage and/or erythrophagocytosis in the mesenteric lymph node was present in rat #33068 (100 mg/kg).

Although gastro-intestinal lesions were not found histopathologically in sacrificed rats, necrosis, hemorrhage, and acute inflammation were observed in stomach of rat #33062 (50 mg/kg), a rat previously noted as having a reddened glandular stomach. This rat also

had slight intestinal epithelial necrosis. Rat #33068 (100 mg/kg) had slight mucosal hemorrhages in both the stomach and small intestine.

3. Summary.

a. The low numbers of deaths reflect the relative innocuous nature of 4-Nitrophenyl Monochloromethyl (phenyl) phosphinate when given at the dose levels of 12.5, 25, 50, and 100 mg/kg by gavage in a Tween 80 based vehicle for fourteen days.

b. The deaths of one 12.5 and one 100 mg/kg rat could be attributed to the gavage procedure and hence were not directly compound related. The unscheduled deaths of the other two rats (50 and 100 mg/kg), however, may have been due to the toxic effects of the compound and, in these cases, were specifically manifested by gross and microscopic gastro-intestinal irritation.

c. Gross necropsy observations in sacrificed animals from all treatment groups revealed no compound related effects. Similarly there were no distinct compound related histopathologic tissue alterations in these animals. Portally oriented hepatic inflammation, renal tubular mineralizations, and hemorrhages within lymph nodes were considered to be of dubious significance.

d. The mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration values were decreased in the 50 and 100 mg/kg rats, respectively. Although statistically significant, these differences were not accompanied by other statistically significant hematologic alterations and are as yet unexplained.

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13 November 1987

APPENDICES

I. Appendix A - Supplementary Guide to Interpretation of Histopathologic Observations

II. Appendix B - Key to Tables 1, 2 & 3

- Tables 1, 2 & 3

III. Appendix C - Statistical Analysis of Hematologic Values, Study #82-034

APPENDIX A

Supplementary Guide to Interpretation of Histopathologic Observations

The following observations were not coded as they occur with considerable frequency in normal male Sprague Dawley Rats.

1. Interstitial, paraductular lymphoid aggregates in the pancreas and salivary glands.

2. Plasmacytosis and lymphoid hyperplasia of very slight degrees in the submandibular lymph node.

3. Very slight to slight hemosiderin deposition in the spleen.

4. Very slight degrees of sinus ectasia, sinus histiocytosis, and lymphoid hyperplasia in the mesenteric lymph node. Greater degrees were coded.

5. Submucosal lymphoid aggregates in nasal cavity. Acute inflammation in paired vomeronasal organs. Flocculent eosinophilic material +/- artifactually induced hemorrhage within lumens of sinuses.

6. Very slight lymphoid aggregates in seminal vesicles or prostate.

7. Tiny inconspicuous foci of mineralization in gastric glandular epithelium. Very slight aggregates of neutrophils, lymphocytes, and other inflammatory cells in the submucosa and lamina propria of the stomach.

8. Artifactual vacuolation of neurons and white matter of brain and/or spinal cord.

9. Slight amounts of flocculent eosinophilic material within the middle ear.

Very slight progressive nephropathy diagnosed in the kidney when there was evidence of early glomerular alterations (capsular basement thickening $\pm$ synechia and hypercellularity) $\pm$ tubular epithelial hyperplasia and the variable presence of local inflammatory cell infiltrates.

Subacute hepatitis was used to describe foci of lymphoid cells accompanied by cellular degeneration and/or necrosis and the variable presence of neutrophils and/or macrophages.

APPENDIX B

Fourteen Day Sub-chronic Toxicity Study of 4-Nitrophenyl Monochloromethyl(phenyl)phosphinate in Male Albino Sprague-Dawley Rats, Study 82034

Key to Microscopic Findings (Tables 1 - 3):

1. (+) = Tissue or organ present, no significant lesions were observed unless recorded as present (P) or graded as to severity (1-5).

2. (-) = Tissue or organ not present.

3. (P) = Lesion recorded as present and not graded as to severity.

4. Grading for severity of lesion is as follows:

1 = minimal

2 = mild

3 = moderate

4 = marked

5 = severe.

5. ([]) = Gross lesions observed during necropsy.

6. (*) = No gross lesions.

7. Died (x)/Moribund (m) = Rats that died during the study or were killed when moribund.

TABLE #1

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
Died (x)/Moribund (m)			x
No Gross Lesions (*)	* * * * * * * * * *	* * * * * * * * *	* * * * * * *
BRAIN	+ + + + + + + + + +	+ + + + + + + + + +	
[soft]			
TRACHEA	+ + + + + +	+ - + + + + +	
Submucosal lymphoid aggregates	2 2 2 1 2	1	
Subacute tracheitis	2	1	
Peritracheal hemorrhage			
THYROID	+ + + + + + +	+ + - + + + - + + +	
Cyst(s) with keratinaceous debris	1 1 1 1		
PARATHYROID	+ - - - - + - - - +	- - - - - - - + - -	

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
ESOPHAGUS	+ + + + + + + + + +	+ + + + + + + + -	
[dark red subserosal focus]			
Chronic periesophageal inflammation			
Acute esophagitis, periesophagitis			
SALIVARY GLANDS	+ + + + + + + + + +	+ + + + + + + + + +	
EXORBITAL LACRIMAL GLAND	+ + + + + + + + +	+ + + + + + + + + +	
Subacute adenitis	2 3		
HARDERIAN GLAND	+ + + + + + + + + +	+ + + + + + + + + +	+ + + + + + + + + +
Subacute adenitis			
HEART	+ + + + + + +	+ + + + + + +	+ + + + + + +
Endocarditis, subacute, nonsuppurative	1		
Epicarditis, nonsuppurative	1	1	
Myocarditis, nonsuppurative	1	1 1	1
Lymphoid aggregates	1	1	1 1

APPENDIX G-1 (cont.)

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
ACCESSION #	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
	0 0 0 0 0 0 0 0 1 1	0 0 0 0 0 0 0 0 0 1	0 0 0 0 0 0 0 0 1 1
	6 7 7 7 8 8 9 9 0 1	5 5 6 7 7 7 8 8 9 0	6 6 6 8 8 8 9 9 0 1
	4 0 1 7 2 5 6 8 7 3	7 9 7 2 4 8 3 6 9 1	0 1 3 0 4 7 2 5 4 1
LUNGS		+	
[yellow brown subpleural focus]			
[red brown foci]			
[firm dark non collapsed lobes plus oily material in thorax]			
[red brown mottling]			
Bronchiolitis and peribronchiolitis, subacute	2		
Subpleural lymphoid aggregates	1 1		
Parabronchial lymphoid aggregates	1 1 1 2 2 2 1 1 2 2	2 2 2 1 1 1 2 2 1	
Paravascular lymphoid aggregates	1 1 1	1	
Alveolar hemorrhage	1 1 2 1 2 2 2	1 1 1 1	
Alveolar histiocytosis	1		

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
LUNGS (continued)			
Interstitial pneumonitis, subacute to acute		1 1	
Pleural fibrosis		1 1	
Hemoglobin crystals in alveoli			
Congestion			
MESENTERIC LYMPH NODE	+ + + + + + + + + +	+ + + + + + + + +	
Lymphoid hyperplasia		2	
Hemorrhage and/or erythrophagocytosis			
SUBMANDIBULAR LYMPH NODE	+ - + + + + + + + +	+ - + + - - + - + +	
THYMUS	+ + + + + + + + + +	+ + + + + + + + + +	
Hemorrhage			
SPLEEN	+ + + + + + + + + +	+ + + + + + + + + +	
[thickening of splenic capsule]			P
Subacute splenitis, capsulitis, pericapsulitis/splenitis			2

APPENDIX G-1 (cont.)

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY / [illegible]SION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
NASAL CAVITY/SINUSES	- +	+ + + + + +	
Sinusitis, subacute, maxillary	2 1 2 1 2 2 1	2 1 1 1	
LIVER	+ +		
Acute hepatitis		2	
Biliary hyperplasia			
Periportal subacute hepatitis			
Hepatitis, subacute, random	1 1 1 1 1 1 1	1 1 1 1 1 1 1 1 1	1 1 1 1 1 1 1
Individual cell necrosis	1		
Aggregates of mononuclear cells, primarily lymphoid	1 1 1 1 1 1 1 1	1 1 1 1 1 1 1 1 1 1	1 1 1 1 1 1 1 1
Midzonal hepatocellular vacuolation			
Necrosis			

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B										Vehicle Control 2A & 2B										12.5 mg/kg/day 3A & 3B									
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3										3 3 3 3 3 3 3 3 3 3										3 3 3 3 3 3 3 3 3 3									
	3 3 3 3 3 3 3 3 3 3										3 3 3 3 3 3 3 3 3 3										3 3 3 3 3 3 3 3 3 3									
	0 0 0 0 0 0 0 0 1 1										0 0 0 0 0 0 0 0 0 1										0 0 0 0 0 0 0 0 1 1									
	6 7 7 7 8 8 9 9 0 1										5 5 6 7 7 7 8 8 9 0										6 6 6 8 8 8 9 9 0 1									
	4 0 1 7 2 5 6 8 7 3										7 9 7 2 4 8 3 6 9 1										0 1 3 0 4 7 2 5 4 1									
KIDNEYS		+									+	+				+		+											+	
[dilated renal pelvis - unil.]																	P													
Dilated pelvis (uni or bilateral)			2		2		2	2	2				2		1			2												2	
Mineralization tubules	1												1								1		1	1			1	1		1
Interstitial nephritis, subacute	1			2		2		1		1												1	1		1		1	1		
Progressive nephropathy	1					1							1													1	1	1		
Tubular epithelial hyperplasia				2				1		2				1	1		1		1			1	1	1	1					1
Basophilic tubules																														
URINARY BLADDER	+	+	+	+	+	+	+	+	+		+	+	+	+	+	+	+	+	+	+										
Lymphoid aggregates										1																				

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
PROSTATE	+ + + + + + + + +	+ + + + + + + + + +	
Subacute prostatitis (confined to glandular lumen)	1		
Lymphoid aggregates increased			
ACCESSORY SEX GLANDS	+ + + + + + + + +	+ + + + + + + + + +	
Acute inflammation ductus deferens	1		
SEMINAL VESICLES	+ + + + + + + + + +	+ + + + + + + + + +	
TESTES	+ + + + + + + + + +	+ + + + + + + + +	
Tubular degeneration		1	
STOMACH	+ + + + + + + + + +	+ + + + + + + + +	
[reddened glandular mucosa]			

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
STOMACH (continued)			
[distended]			
Hemorrhage			
Necrosis - epithelial			
Submucosal - acute inflammation			
Dilated gland(s)		2	
PANCREAS	+ + + + + + + + +	+ + + + + + + + +	
Acute pancreatitis	1	1	
SMALL INTESTINE	+ + + + + + + + + +	+ + + + + + + + + +	
[distended with mucoid material]			
Necrosis - epithelial			
Hemorrhage			

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
CECUM	+ + + + + + + + + +	+ + + + + + + + + +	
COLON	+ + + + + + + + + +	+ + + + + + + + + +	
SKELETAL MUSCLE	+ + + + + + + + + +	+ + + + + + + + + +	
SCIATIC NERVE	+ + + + + + + + + +	+ + + + + + + + + +	
SKIN	+ + + + + + + + + +	+ + + + + + + + + +	
[focal abrasion]			P
[oily red material at muzzle]			P
VERTEBRAE	+ + + + + + + + + +	+ + + + + + + + + +	
SPINAL CORD	+ + + + + + + + + +	+ + + + + + + + + +	
RIB	+ + + + + + + + + +	+ + + + + + + + + +	

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	Cage Control 1A & 1B	Vehicle Control 2A & 2B	12.5 mg/kg/day 3A & 3B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 7 7 7 8 8 9 9 0 1 4 0 1 7 2 5 6 8 7 3	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 0 1 5 5 6 7 7 7 8 8 9 0 7 9 7 2 4 8 3 6 9 1	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 6 6 6 8 8 8 9 9 0 1 0 1 3 0 4 7 2 5 4 1
FEMUR	+ + + + + + + + - +	+ + + + + + + + + +	
ADRENALS	+ + + + + + + + + +	+ + + + + + + + + +	
Sinusoidal ectasia			
PITUITARY	- + - + - - + +	+ + + + - + + + + +	
Cyst, microscopic	1		
Aggregates of lymphoid cells	1		
EYES	+ + + + + + + + + +	+ + + + + + + + + +	
MIDDLE EAR	+ + - + + + + + + +	+ + + + + + + + + +	

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
Died (x)/Moribund (m)		x	x x
No Gross Lesions (*)	* * * * * * * * *	* * * * * * * *	* * * * * * *
BRAIN			+ + + + + + + + +
[soft]			P
TRACHEA			- + + + + + -
Submucosal lymphoid aggregates			1 2
Subacute tracheitis			
Peritracheal hemorrhage			2
THYROID			- + + + + + + + -
Cyst(s) with keratinaceous debris			1
PARATHYROID			- - - - - - - - - -

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL	25/mg/kg/day	50 mg/kg/day	100 mg/kg/day
GROUP #	4A & 4B	5A & 5B	6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
ESOPHAGUS			- + + + + + + +
[dark red subserosal focus]			P
Chronic periesophageal inflammation			3
Acute esophagitis, periesophagitis			3
SALIVARY GLANDS			+ + + + + + + + + +
EXORBITAL LACRIMAL GLAND			+ + + + + + + + + +
Subacute adenitis			
HARDERIAN GLAND			+ + + + + + + + + +
Subacute adenitis			1
HEART	+ + + + + + + + + + + + + + + + +		+ + + + +
Endocarditis, subacute, nonsuppurative			
Epicarditis, nonsuppurative			2 1
Myocarditis, nonsuppurative	1 1 1		1 1
Lymphoid aggregates	1 1 1 1	1	1 1 1 1

APPENDIX G-1 (cont.)

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
LUNGS			
[yellow brown subpleural focus]	P		
[red brown foci]		P	
[firm dark noncollapsed lobes plus oily material in thorax]			P
[red brown mottling]			P
Bronchiolitis and peribronchiolitis, subacute			
Subpleural lymphoid aggregates			
Parabronchial lymphoid aggregates		1	1 2 2 1 1 1 1 1 1
Paravascular lymphoid aggregates			1 1 1 1
Alveolar hemorrhage		2	2 1 1 2
Alveolar histiocytosis			1

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 4 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
LUNGS (continued)			
Interstitial pneumonitis, acute to subacute		2	1 1 1
Pleural fibrosis			
Hemoglobin crystals in alveoli		2	
Congestion			3
MESENTERIC LYMPH NODE		+	+ + + +
Lymphoid hyperplasia			2
Hemorrhage and/or erythrophagocytosis			2 1 2 2 1
SUBMANDIBULAR LYMPH NODE			+ - + + + + - + - -
THYMUS			+ + + + + + + + + +
Hemorrhage			2
SPLEEN			+ + + + + + + + + +
[thickening of splenic capsule]			
Subacute splenitis, capsulitis, pericapsulitis/splenitis			

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
NASAL CAVITY/SINUSES			+ + + + +
Sinusitis, subacute, maxillary			2 1 2 2 1
LIVER			
Acute hepatitis			2
Biliary hyperplasia		2	
Periportal subacute hepatitis		2 2 2	3 2 2 3
Hepatitis, subacute, random	1 1 1 1 1 1 1 1 1 1	1 1 2 1 1 1 1	1 1 1 1 1
Individual cell necrosis		2	
Aggregates of mononuclear cells, primarily lymphoid	1 1 1 1 1 1 1 1	1 1 1 1 1 1	1 1 1 1 1 1 1 1 1
Midzonal hepatocellular vacuolation	1		1
Necrosis		2	

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
KIDNEYS	+ + +	+	+ +
[dilated renal pelvis - unil.]			P
Dilated pelvis (uni or bilateral)	2	2 2	3 2 2
Mineralization tubules	1 1 2 1 1 1	1 1 1	1 2 1 2 2
Interstitial nephritis, subacute	1 1 1	1 1 1 2	1 2
Progressive nephropathy	1	1 1 1	1
Tubular epithelial hyperplasia	1 1	2 1 1 1	1 2 3
Basophilic tubules	1		
URINARY BLADDER			+ + + + + + + + + +
Lymphoid aggregates			

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL / GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 1 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
PROSTATE			+ + + + + + + + +
Subacute prostatitis (confined to glandular lumen)			
Lymphoid aggregates increased			2
ACCESSORY SEX GLANDS			+ + + + + + + + + +
Acute inflammation ductus deferens			
SEMINAL VESICLES			+ + + + + + + + + +
TESTES			+ + + + + + + + +
Tubular degeneration			1 3
STOMACH			+ + + + + + + +
[reddened glandular mucosa]		P	P

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL GROUP #	25/mg/kg/day 4A & 4B	50 mg/kg/day 5A & 5B	100 mg/kg/day 6A & 6B
LAIR PATHOLOGY ACCESSION #	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 7 7 7 8 8 9 0 0 0 4 3 5 6 1 9 7 0 5 9	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 1 1 1 5 6 6 7 8 9 0 0 0 1 6 2 9 9 8 4 2 6 8 0	3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 3 0 0 0 0 0 0 0 0 1 1 5 5 6 6 6 9 9 9 0 1 5 8 5 6 8 0 1 3 3 2
STOMACH (continued)			
[distended]			P
Hemorrhage		2	2
Necrosis - epithelial		2	
Submucosal - acute inflammation		2	2
Dilated gland(s)			
PANCREAS			+ + + + + + + + + +
Acute pancreatitis			
SMALL INTESTINE			+ + + + + + + + + +
[distended with mucoid material]		P	P
Necrosis - epithelial		2	
Hemorrhage			2

APPENDIX G-1 (cont.)

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL	25/mg/kg/day	50 mg/kg/day	100 mg/kg/day
GROUP #	4A & 4B	5A & 5B	6A & 6B
LAIR PATHOLOGY	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
ACCESSION #	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
	0 0 0 0 0 0 0 1 1 1	0 0 0 0 0 0 1 1 1 1	0 0 0 0 0 0 0 0 1 1
	5 7 7 7 8 8 9 0 0 0	5 6 6 7 8 9 0 0 0 1	5 5 6 6 6 9 9 9 0 1
	4 3 5 6 1 9 7 0 5 9	6 2 9 9 8 4 2 6 8 0	5 8 5 6 8 0 1 3 3 2
CECUM			+ + + + + + + + + +
COLON			+ + + + + + + + + +
SKELETAL MUSCLE			+ + + + + + + + + +
SCIATIC NERVE			+ + + + + + + + + +
SKIN			+ + + + + + + + + +
[focal abrasion]			
[oily red material at muzzle]			P
VERTEBRAE			+ + + + + - + + + +
SPINAL CORD			+ + + + + - + + + +
RIB			+ + + + + + + + + +

TABLE #1 (continued)

Summary of Individual Gross and Microscopic Findings in Male Sprague Dawley Rats,
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

DOSAGE LEVEL	25/mg/kg/day	50 mg/kg/day	100 mg/kg/day
GROUP #	4A & 4B	5A & 5B	6A & 6B
LAIR PATHOLOGY	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
ACCESSION #	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3	3 3 3 3 3 3 3 3 3 3
	0 0 0 0 0 0 0 1 1 1	0 0 0 0 0 0 1 1 1 1	0 0 0 0 0 0 0 0 1 1
	5 7 7 7 8 8 9 0 0 0	5 6 6 7 8 9 0 0 0 1	5 5 6 6 6 9 9 9 0 1
	4 3 5 6 1 9 7 0 5 9	6 2 9 9 8 4 2 6 8 0	5 8 5 6 8 0 1 3 3 2
TETUR			+ + + + + + + + + +
BONE MARROW			+ + + + + + + + + +
ADRENALS			+ + + + + + + + + +
Sinusoidal ectasia			2
PITUITARY			+ + + + + + + + + +
Cyst, microscopic			
Aggregates of lymphoid cells			
EYES			+ + + + + + + + + +
MIDDLE EAR			+ + + + + + + + + +

TABLE #2

Group Summary of Gross Necropsy Observations at Termination Sacrifice in Male Sprague Dawley Rats
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
Number of animals examined:	10	10	9	10	9	8
Number of animals with no gross lesions	10	9	7	9	8	7
LUNGS						
Yellow brown subpleural focus	0*	0	0	1	0	0
Red brown foci	0	0	0	0	1	0
SPLEEN						
Thickening in splenic capsule	0	0	1	0	0	0
KIDNEY						
Dilated renal pelvis (uni or bilateral)	0	1	0	0	0	1
SKIN						
Focal abrasion	0	0	1	0	0	0

*Number of rats in each group with gross lesions.

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
TRACHEA	10*	9	0	0	0	6
Tracheitis	1**	1				0
Lymphoid aggregates	4	1				1
Peritracheal hemorrhage	0	0				1
THYROID	10	8	0	0	0	6
Cysts with keratinaceous debris	4	0				1
ESOPHAGUS	10	9	0	0	0	7
Chronic periesophagitis	0	0				1
Acute esophagitis and periesophagitis	0	0				1
EXTRAORBITAL LACRIMAL GLAND	10	0	0	0	0	8
Adenitis	2					0

*Number of tissues/organs examined microscopically.
**Number of tissues/organs examined microscopically with the lesions

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
HARDERIAN GLAND	10	0	0	0	0	8
Adenitis	0					1
HEART	10	10	9	10	9	8
Epicarditis	1	1	0	0	0	2
Myocarditis	1	2	1	3	0	2
Endocarditis	1	0	0	0	0	0
Lymphoid aggregates	1	1	2	4	1	4
LUNGS	10	10	0	0	1	8
Bronchiolitis/peribronchiolitis	1	0			0	0
Subpleural lymphoid aggregates	2	0			0	0
Perivascular lymphoid aggregates	3	1			0	4
Parabronchial lymphoid aggregates	10	9			1	8
Hemorrhage	7	4			1	2

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
LUNGS (continued)	10	10	0	0	1	8
Interstitial pneumonitis	0	2			1	2
Pleural fibrosis	0	2			0	0
Alveolar histiocytosis	1	0			0	1
Hemoglobin crystals	0	0			1	0
MESENTERIC LYMPH NODE	10	10			1	8
Lymphoid hyperplasia	0	1			0	1
Hemorrhage and/or erythrophagotosis	0	0			0	5
SPLEEN	10	10	1	0	0	8
Splenitis, capsulitis, perisplenitis	0	0	1			0
NASAL CAVITY	10	10	0	0	0	8
Sinusitis	7	4				3

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
LIVER	10	10	9	10	9	8
Biliary hyperplasia	0	0	0	0	1	0
Periportal hepatitis - subacute	0	0	0	0	2	3
Acute random hepatitis	0	1	0	0	0	1
Subacute random hepatitis	7	9	7	10	7	4
Individual cell necrosis	1	0	0	0	0	0
Aggregates of mononuclear cells	8	10	8	10	6	4
Midzonal hepatocellular vacuolation	0	0	0	1	0	1
Necrosis	0	0	0	0	0	0
KIDNEY	10	10	9	10	9	8
Dilated pelvis	5	3	1	1	2	3
Tubular mineralization	1	1	6	6	3	3

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (LAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
KIDNEY (continued)	10	10	9	10	9	8
Interstitial nephritis	5	0	4	3	3	2
Progressive nephropathy	2	1	3	1	3	1
Tubular epithelial hyperplasia	3	4	4	2	4	2
Basophilic tubules	0	0	0	1	0	0
URINARY BLADDER	10	10	0	0	0	8
Lymphoid aggregates	1	0	0	0	0	0
PROSTATE	10	10	0	0	0	8
Prostatitis	1	0				0
Lymphoid aggregates	0	0				1
DUCTUS DEFERENS	10	10	0	0	0	8
Acute inflammation	1	0				0

TABLE #3

Group Summary of Histopathologic Observations on Male Sprague Dawley Rats Surviving to Terminal Sacrifice
GLP Study #82034
14 Day Subchronic Oral Toxicity Study for 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate (IAIR TA009)

Tissue/Organ	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6
Dosage level:	Cage Control	Vehicle Control	12.5 mg/kg	25 mg/kg	50 mg/kg	100 mg/kg
TESTES	10	10	0	0	0	8
Seminiferous tubular degeneration	0	1				2
STOMACH	10	10	0	0	0	8
Dilated glands	0	1				0
Submucosal inflammation	0	0				1
PANCREAS	10	10	0	0	0	8
Pancreatitis	1	1				0
ADRENALS	10	10	0	0	0	8
Sinusoidal ectasia	0	0				1
PITUITARY	4	9	0	0	0	8
Lymphoid aggregates	0	1				0
Cyst	0	1				0

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

REPLY TO
ATTENTION OF:
SGRD-ULZ-I

6 June 1985

MEMORANDUM FOR RECORD

SUBJECT: Statistical Analysis/Study #82-034

1. A computer package, BMDP on the Data General MV8000 computer, was utilized to analyze the hematology data of study #82-034.

2. Student's t-tests were performed to compare the measurements of the cage control group with the vehicle control group. No significant differences between the groups were found for red blood cell count, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin concentration, mean corpuscular hemoglobin, reticulocytes, platelets, white blood cell count, neutrophils, lymphocytes, eosinophils and monocytes values.

3. One-way analysis of variance was used to test for differences among the vehicle control, 12.5 mg/kg, 25 mg/kg, 50 mg/kg, and 100 mg/kg dose groups. When a significant F-value for a group effect was found, a posteriori multiple comparisons were used to test for differences among means for the vehicle control group with a one-sided Dunnett's test.

4. No differences were found for the following: red blood cell count, hemoglobin, mean corpuscular volume, reticulocytes, platelets, white blood cell count, neutrophils, lymphocytes, eosinophils and monocytes.

5. The dose group, 12.5 mg/kg, was found to have a significantly greater mean for the hematocrit than the vehicle control. The mean corpuscular hemoglobin mean value was found to be significantly lower for the 50 mg/kg group than the vehicle control group. In addition, the 100 mg/kg group had a significantly lower mean for the mean corpuscular hemoglobin concentration values than the vehicle control group.

6. The 0.05 level of significance was used with all statistical tests.

Virginia L Gildengorin

VIRGINIA L. GILDENGORIN, PhD
Chief, Biometric Team, ISG

14 Day Subchronic Oral Toxicity in Male Rats
of 4-Nitrophenyl Monochloromethyl (Phenyl) Phosphinate

INDIVIDUAL HEMATOLOGY DATA

Group	Animal ID	RBC Count	HGB	HCT	MCV	MCH	MCHC	Reticulocytes	Platelets	WBC Count	WBC Differential Count - Absolute #			
											Neutrophils	Lymphocytes	Eosinophils	Monocytes
		6 x10 /ul	g/dl	%	3 u	uug	%	%	6 x10/ul	3 x10 /ul	3 x10 /ul	3 x10 /ul	3 x10 /ul	3 x10 /ul
1	82D00986	7.62	16.2	44.0	61	21.2	36.8	3.2	864	4.9	0.7	4.1	0.0	0.0
	82D00992	7.56	17.0	44.9	62	22.5	37.9	3.0	1253	9.8	1.9	7.7	0.0	0.0
	82D00993	7.63	17.1	44.5	61	22.4	38.4	2.9	915	8.1	0.6	7.4	0.0	0.0
	82D01000	7.21	17.5	41.6	61	24.3	42.1	1.9	973	8.9	0.7	8.1	0.0	0.0
	82D01007	7.29	16.4	39.0	56	22.5	42.1	2.9	736	7.8	0.7	7.0	0.0	0.0
	82D01010	8.02	15.8	44.4	58	19.7	35.6	1.9	973	7.0	2.0	4.9	0.0	0.0
	82D01022	7.78	16.8	45.0	61	21.6	37.3	2.3	817	6.3	0.8	5.4	0.0	0.0
	82D01025	8.70	16.9	46.8	57	19.4	36.1	2.9	813	6.8	0.8	5.7	0.0	0.1
	82D01035	8.60	17.8	42.9	53	20.7	41.5	3.7	654	9.5	1.5	7.7	0.0	0.1
	82D01041	7.62	17.6	44.9	62	23.1	39.2	3.9	785	8.5	1.2	7.1	0.0	0.0
2	82D00977	7.45	15.7	38.3	54	21.2	40.8	2.9	815	5.8	0.6	4.8	0.1	0.1
	82D00980	7.51	15.9	45.9	64	21.2	34.6	3.0	1153	6.5	1.3	5.1	0.0	0.0
	82D00989	7.76	17.1	45.1	61	22.0	37.9	3.5	886	7.7	1.4	6.2	0.0	0.0
	82D00994	7.31	16.1	41.1	59	22.0	39.2	1.7	893	7.1	0.8	6.1	0.0	0.0
	82D00997	7.96	17.0	41.4	55	21.4	41.1	2.8	800	8.6	0.7	7.7	0.0	0.0
	82D01001	7.70	16.6	40.5	55	21.6	41.0	2.1	783	8.7	1.2	7.4	0.0	0.0
	82D01008	8.25	16.9	42.1	54	20.5	40.1	2.9	863	8.5	1.5	6.9	0.0	0.0
	82D01011	7.39	16.1	40.6	58	21.8	39.7	4.3	717	6.9	0.6	5.1	0.1	0.0
	82D01026	7.61	17.4	43.1	60	22.9	40.4	2.9	723	7.2	0.5	6.6	0.0	0.0
	82D01028	7.27	17.0	40.7	59	23.4	41.8	3.1	913	10.4	1.2	9.1	0.0	0.0
3	82D00981	7.53	17.0	42.1	59	22.7	40.4	3.4	671	7.4	1.7	5.6	0.0	0.0
	82D00983	8.04	17.5	45.2	59	21.8	38.7	2.8	825	9.1	1.9	7.0	0.0	0.1
	82D00985	7.36	16.0	41.6	59	21.7	38.5	2.8	1070	7.6	0.9	6.5	0.0	0.1
	82D01004	8.62	16.0	47.5	58	18.6	33.7	3.9	834	6.4	0.6	5.5	0.1	0.0
	82D01012	8.54	17.5	46.4	57	20.5	37.7	2.7	1093	6.0	0.6	5.2	0.0	0.0
	82D01018	7.33	16.2	42.3	61	22.1	38.3	3.0	1183	7.3	0.6	6.6	0.0	0.0
	82D01021	8.56	17.8	47.9	59	20.8	37.2	4.1	844	7.5	0.9	6.5	0.0	0.0
	82D01032	8.70	17.9	48.2	58	20.6	37.1	3.1	883	7.0	1.0	5.8	0.1	0.0
	82D01039	8.18	17.4	46.6	60	21.2	37.3	3.4	713	8.0	0.7	7.2	0.0	0.0

INDIVIDUAL HEMATOLOGY DATA

Group	Animal ID	RBC Count	HGB	HCT	MCV	MCH	MCHC	Reticulo-cytes	Plate-lets	WBC Count	WBC Differential Count - Absolute #			
											Neutro-phils	Lympho-cytes	Eosino-phils	Mono-cytes
		6			3				6	3	3	3	3	3
		x10 /ul	g/dl	%	u	uug	%	%	x10/ul	x10 /ul	x10 /ul	x10 /ul	x10 /ul	x10 /ul
4	82D00974	8.10	16.8	41.2	53	20.7	40.8	1.9	715	6.4	0.7	5.5	0.0	0.0
	82D00996	6.42	14.6	38.1	63	22.7	38.3	4.0	807	8.5	0.9	7.0	0.1	0.3
	82D00998	8.16	16.9	44.6	57	20.7	37.9	2.4	943	6.7	0.6	6.0	0.0	0.0
	82D00999	8.08	17.8	45.8	60	22.0	38.9	4.0	922	6.3	0.6	5.5	0.0	0.0
	82D01005	7.08	16.1	37.4	56	22.7	43.0	2.0	753	8.1	0.4	7.6	0.0	0.0
	82D01014	7.81	16.5	46.2	62	21.2	35.7	2.9	1239	5.6	0.6	4.9	0.0	0.0
	82D01023	8.20	17.3	45.3	58	21.1	38.2	3.7	841	5.8	1.0	4.7	0.0	0.0
	82D01027	8.09	17.4	42.2	55	21.5	41.2	3.4	690	6.4	0.7	5.6	0.0	0.0
	82D01033	8.16	17.9	42.5	55	21.9	42.1	4.2	768	11.4	0.4	10.9	0.0	0.0
	82D01037	7.93	17.4	40.2	53	21.9	43.3	4.0	683	7.8	0.8	6.9	0.0	0.0
5	82D00976	7.81	15.3	44.6	60	19.6	34.3	2.2	1008	6.2	0.8	5.2	0.0	0.0
	82D00991	7.33	15.8	41.7	60	21.6	37.9	3.1	905	6.5	1.3	5.0	0.1	0.0
	82D01003	7.58	15.6	39.9	53	20.6	39.1	3.7	797	9.0	0.9	7.8	0.2	0.0
	82D01013	8.67	16.5	45.4	55	[illegible].0	36.3	4.0	827	7.0	1.4	5.4	0.0	0.0
	82D01020	7.97	17.0	46.9	62	2[illegible].3	36.2	4.0	1035	5.9	0.8	4.8	0.1	0.0
	82D01029	8.16	16.0	43.6	56	19.6	36.7	3.6	776	7.0	0.8	6.0	0.0	0.0
	82D01034	8.06	16.9	41.6	54	21.0	40.6	3.9	724	7.4	0.6	6.7	0.0	0.0
	82D01036	8.85	17.8	43.0	51	20.1	41.4	3.8	653	7.2	0.9	6.2	0.0	0.0
	82D01038	8.06	17.4	44.0	57	21.6	39.5	3.6	1037	9.3	0.7	8.4	0.0	0.0
6	82D00975	8.01	15.3	44.7	59	19.1	34.2	4.5	563	7.7	2.7	4.8	0.0	0.0
	82D00979	7.13	15.0	39.5	59	21.1	38.0	3.0	1200	7.7	0.9	6.7	0.0	0.0
	82D00987	7.67	16.4	43.6	60	21.4	37.6	4.0	973	7.2	0.7	6.4	0.0	0.0
	82D00988	7.05	14.5	40.2	60	20.6	36.1	3.9	770	6.8	0.9	5.7	0.0	0.1
	82D01015	7.94	17.6	44.3	59	22.2	39.7	4.0	1001	5.7	0.5	5.1	0.0	0.0
	82D01017	7.67	17.3	44.5	61	22.6	38.9	3.8	882	8.2	1.0	7.0	0.0	0.0
	82D01030	8.31	16.4	45.2	57	19.7	36.3	3.6	809	7.2	0.9	6.0	0.2	0.0
	82D01040	7.99	17.0	44.7	59	21.3	38.0	3.7	727	7.8	1.2	6.5	0.0	0.0

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