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# Effects of subclinical lead exposure on the resistance of swine to challenge with Salmonella choleraesuis var Kunzendorf

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# Effects of subclinical lead exposure on the resistance of swine to challenge with *Salmonella choleraesuis* var. *Kunzendorf*

by

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# INTRODUCTION

In work performed in the Toxicology Section, Iowa Veterinary Diagnostic Laboratory and the Department of Veterinary Microbiology and Preventive Medicine, Iowa State University, Hemphill (1973) found that mice orally exposed to low levels of lead nitrate experienced significantly higher mortality rates than non-lead-exposed controls when challenged with virulent Salmonella typhimurium organisms. These studies also revealed that lead nitrate-exposed mice which were inoculated with a bacterin of S. typhimurium developed significantly lower agglutinating antibody titers and had significantly lower serum $alpha_1$ and gamma globulin concentrations than did non-lead-exposed mice.

In light of the ubiquitous natural distribution of lead (Chisolm, 1971) and the resultant exposure of all animals, including man, to this metal, the finding of both lead-induced reduction of resistance to infection and alteration of certain immunologic parameters has important biological implications. The effect of lead exposure on immunity and/or resistance should, therefore, be studied in another mammalian species more closely related to man. The pig is physiologically similar to man (Earl et al., 1964; Bustad and McClellan, 1966) and should be a useful test animal.

The purpose of this investigation was to determine the effect of subclinical levels of lead exposure on the

resistance of swine to bacterial infection. It was proposed that such an effect might be detected by observing relative mortality rates in lead- and non-lead-exposed pigs subsequently challenged with a gram-negative, endotoxin-synthesizing bacteria. Furthermore, it was felt that measurement of physiologic parameters such as serum antibody titers against bacterial antigens, serum protein electrophoretic patterns (with special attention to relative globulin concentrations), total leukocyte counts, and leukocyte differential counts could give valuable information on the response of the animals to such a challenge.

It was also hoped that such studies would reveal any other physiopathologic alterations caused by lead exposure in swine.

# LITERATURE REVIEW

Excellent reviews concerning sources and extent of environmental lead contamination (Patterson, 1965), metabolism of lead in the animal body (Kehoe, 1961), and possible adverse effects of lead on health (Chisolm, 1971; De Bruin, 1971) have previously been compiled.

Chisolm (1971) stated that "Among the natural substances that man concentrates in his immediate environment, lead is one of the most ubiquitous," and Patterson (1965) contended that "the average resident of the United States is being subjected to severe chronic lead insult." It is evident that lead wastes have been accumulating during the last century and that lead concentrations in organs of individuals from highly industrialized societies are higher than those of most individuals from primitive populations (Chisolm, 1971).

The best known adverse effect of lead on the living organism is inhibition of the activities of enzymes which are dependent on the presence of free sulfhydryl groups for their activities (Chisolm, 1971). The classic example of such an effect is lead-induced inhibition of activities of enzymes in the heme synthetic pathway.

Lead is thought to inhibit energy metabolism of certain cells of the kidney and, after long-term exposure, to cause chronic kidney failure (Chisolm, 1971; De Bruin, 1971).

Lead adversely affects the central nervous system through upset of energy metabolism and/or direct injury to neurons, and also injures the peripheral nervous system by causing nerve sheath demyelination (Chisolm, 1971).

It has been theorized that lead reduces thyroid gland activity, efficiency of the clotting mechanism, and immunologic responsiveness (De Bruin, 1971).

The concept of immunosuppression caused by environmental contaminants (such as lead) is not new. A number of workers have investigated the potential immunosuppressive properties of such contaminants as polychlorinated biphenyls (PCBs), particulate carbon, sulfur dioxide, cobalt, arsenicals, cadmium, mercury, and lead.

Friend and Trainer (1970) noted that mallard ducklings orally exposed to PCBs and then challenged with duck hepatitis virus suffered more rapid onset of mortality as well as higher mortality rates than did non-PCB-treated controls. Guinea pigs fed low levels of PCBs had significantly decreased antitoxin titers as compared to controls after challenge with tetanus toxoid (Vos and De Roij, 1972; Vos and Van Driel-Grootenhuis, 1972). Such guinea pigs also had decreased numbers of gamma globulin-producing cells in popliteal lymph nodes, decreased serum gamma globulin concentrations, and decreased cell-mediated immune responses as measured by the intradermal tuberculin test (Vos and Van Driel-Grootenhuis,

1972). Koller and Thigpen (1973) found that rabbits fed three different formulations of PCBs developed significantly lower serum-neutralizing antibody titers to pseudorabies virus than did rabbits not fed PCBs. In this same study, serum gamma globulin concentrations were lowered in rabbits receiving one of the PCB formulations but were elevated in rabbits receiving the other two PCB formulations.

Zarkower (1972) observed significant decreases in agglutinin titers in mice exposed to carbon or to a combination of carbon and sulfur dioxide by the aerosol route and then challenged by this same route with Escherichia coli bacterin. Such mice also had decreased numbers of splenic antibody-producing cells. In similar studies using sulfur dioxide without carbon, it was found that there was an increase in agglutinin titers and in numbers of splenic antibody-producing cells after 135 days of exposure but that both decreased after 192 days of sulfur dioxide inhalation.

Mice consuming "non-growth-depressing" concentrations of cobalt sulfate had significantly higher mortality rates and virus titers in brain, heart, and spleen than did controls after exposure to encephalomyocarditis virus (Gainer, 1972b). These studies also revealed that cobalt sulfate inhibited the action of interferon in cell cultures.

Berry and Mitchell (1953) reported increased mortality rates in mice parenterally exposed to sodium arsenite and

inoculated with Salmonella typhimurium organisms. These workers explained that the increased susceptibility resulted from inhibition of the Krebs cycle and subsequent metabolic upset. Mice treated with sodium arsenite and sodium arsenate suffered significantly higher mortality rates as compared to controls when challenged with pseudorabies virus, encephalomyocarditis virus, or St. Louis encephalitis virus (Gainer and Pry, 1972). These findings were attributed to arsenical-induced inhibition of interferon. In further studies, Gainer (1972a) noted that high levels of exposure to sodium arsenite and roxarsone inhibited both the synthesis and action of interferon in vivo and in vitro.

Jones et al. (1971) reported that rats receiving intravenous (i.v.) injections of cadmium chloride two weeks prior to initial inoculation with human gamma globulin (HGG) experienced enhanced primary and secondary antibody responses, but that rats receiving similar cadmium injections one week prior to HGG inoculation had delayed primary and depressed secondary antibody responses. Koller (1973) found that rabbits receiving oral cadmium for a 70-day period and then inoculated with formalinized pseudorabies virus developed significantly lower serum-neutralizing antibody titers than did non-cadmium-exposed controls.

Koller (1973) also detected decreased serum-neutralizing antibody titers in mercury-exposed rabbits inoculated with

formalinized pseudorabies virus; however, this decrease was not as marked as that noted in cadmium- or lead-exposed rabbits.

Hemphill (1973) demonstrated that mice orally exposed to lead nitrate and subsequently challenged with virulent S. typhimurium organisms experienced significantly higher mortality rates than did non-lead-exposed controls. In these same studies, mice orally exposed to lead nitrate and then inoculated with a S. typhimurium bacterin developed significantly lower agglutinating antibody titers and had significantly lower serum $alpha_1$ and gamma globulin concentrations as compared to controls. Such mice also had marked decreases in serum properdin activity. Hemphill suggested that this study gave evidence for lead inactivation of antibodies.

Hamsters exposed to different levels of lead for variable periods of time were no more susceptible to S. typhimurium than were non-lead-exposed controls, and no significant alterations in serum protein electrophoretic patterns were detected (Mankus, 1975).

In lead-exposed workers, Soliman et al. (1970) noted decreased serum $alpha_1$ and $alpha_2$ globulin levels, but no decreases in serum gamma globulin concentrations were found. The decreased concentrations of these serum protein fractions were theorized to be related to urinary protein loss.

Rabbits exposed to lead orally for 70 days developed lower serum-neutralizing antibody titers (10X) when challenged with formalinized pseudorabies virus as compared to rabbits not consuming lead (Koller, 1973).

In studying mice inoculated intraperitoneally (i.p.) with sheep red blood cells, Koller and Kovacic (1974) found that those exposed to lead acetate orally had lower numbers of splenic cells producing either IgG or IgM antibodies. These workers felt that chronic lead exposure caused a significant decrease of antibody synthesis and that this probably involved memory cells.

Lead-exposure produced no difference in passive hemagglutination titers in lambs inoculated with Serratia marcescens (Hoffman, 1974). In this same series of studies, lead-exposed lambs had significantly lower serum $alpha_2$ globulin concentrations than did controls, but lead exposure caused no difference in serum IgG or IgM levels.

Vengris and Mare (1974) reported that long-term subclinical lead intake did not suppress either interferon or antibody production in chickens; however, higher levels of lead exposure markedly decreased interferon concentrations and duration of interferon in serum.

Trejo and Di Luzio (1971) found that a single i.v. injection of lead acetate in rats did not modify the primary or secondary immune response to an injection of sheep red

blood cells as measured by the hemagglutination test.

Hoffman et al. (1972) suggested that rather than effecting humoral immunity, lead might act to increase susceptibility to infections by causing impaired phagocytosis or impairment of protein synthesis by phagocytic cells.

There has been extensive study of the relationship of bacterial endotoxin and lead acetate in living organisms. Selye et al. (1966) discovered that rats which received simultaneous i.v. injections of lead acetate and E. coli endotoxin were approximately 100,000 times more sensitive to endotoxin than were rats receiving endotoxin alone. These workers noted that the minimum i.v. dosage of lead acetate needed to induce increased sensitivity to endotoxin was 10 mg/kg body weight.

Other workers have confirmed the endotoxin-sensitizing quality of lead in rats (Filkins, 1970; Filkins and Buchanan, 1973), chicks (Truscott, 1970), mice (Rippe and Berry, 1972; Seyberth et al., 1972) and baboons (Holper et al., 1973).

Trejo and Di Luzio (1971) observed highest mortality rates and most severe tissue damage when rats received simultaneous injections of lead acetate and E. coli endotoxin but noted increased mortalities as compared to rats not exposed to lead acetate when endotoxin was administered from one hour before to seven hours after lead acetate injection. Bertok (1968a) reported that rats rendered endotoxin-tolerant

by a previous small dose of endotoxin experienced a decrease in this tolerance when exposed to a mixture of lead acetate and endotoxin.

In studying the fate of intravenously injected, radio-labeled endotoxin in rabbits, Braude et al. (1955) found that endotoxin was first concentrated in the plasma and then in the buffy coat (especially during leukopenic periods) and the liver. It was felt that the endotoxin accumulation in the liver was probably in phagocytic (Kupffer) cells. These authors also suggested that endotoxin accumulation in neutrophils might cause damage to these cells and subsequent release of pyrogens and other substances mediating endotoxic shock.

Di Luzio and Crafton (1969) demonstrated that i.v.-injected, $Cr^{51}$-labeled S. enteritidis endotoxin was localized in the liver and suggested that this accumulation was mainly in Kupffer cells. Filkins (1970) contended that most detoxification of endotoxin occurred in the liver and spleen. This contention was strengthened by Trejo and Di Luzio (1971) who demonstrated that mice receiving endotoxin which had been previously incubated with homogenates of rat liver and spleen suffered lower mortality rates than did mice receiving endotoxin not previously incubated with such homogenates. These workers did not detect significant endotoxin detoxifying ability in homogenates of lung, kidney, heart, or brain.

Rutenberg et al. (1960) found macrophages to have potent endotoxin detoxifying activity in vitro. Hoffman et al. (1972) felt that Kupffer cells played an important part in uptake and detoxification of endotoxin while hepatocytes were important sites for adjustments of fat, carbohydrate, and protein metabolism necessary for tolerance of endotoxin exposure.

In studies with rabbits previously rendered tolerant to endotoxin, Greisman et al. (1963) found that tolerance could be transferred to other rabbits via plasma and concluded that a humoral factor might play a role in defense against endotoxin. It was suggested that this factor could be an opsonin. Rutenberg et al. (1960) reported that serum from animals which were immune to an endotoxin-producing bacteria had some detoxifying effect on endotoxin from this bacteria. It was also noted that a mixture of macrophages and immune serum detoxified endotoxin more potently than a mixture of macrophages and normal serum.

A single i.v. injection of lead acetate impaired intravascular phagocytosis of colloidal carbon in rats (Trejo et al., 1972; Cook et al., 1974). These workers also demonstrated decreased vascular clearance and decreased hepatic uptake of an R-E test-lipid emulsion following i.v. injection of lead acetate, but no alterations in splenic localization were detected. Trejo et al. (1972) found that

prior opsonization of the R-E test-lipid emulsion did not reverse lead-induced impairment of phagocytosis and concluded that a lack of opsonin was not the mediating factor in such dysfunction. Crafton and Di Luzio (1969) agreed that no stable, circulating plasma factor was involved in altered sensitivity to endotoxin. Selye et al. (1966) compared the endotoxin-sensitizing ability of lead acetate to a variety of reticuloendothelial (R-E) blocking agents and suggested that if lead acetate is an R-E blocking agent, it is more potent than any known.

After an i.v. injection of lead acetate, Hoffman et al. (1972) observed ultrastructural alteration of the mitochondria, endoplasmic reticulum, and lysosomes of Kupffer cells. These cells contained a finely granular, electron-dense material which may have been protein-bound lead. This same material was observed in lysosomes of splenic macrophages. Since these changes occurred later than the period of increased sensitivity to endotoxin, it was felt that they were a sign of earlier functional impairment.

Many workers have cast doubt on the importance of hypophagocytosis as a mechanism causing increased endotoxin sensitivity. Trejo et al. (1972) reported that maximum phagocytic impairment in rats occurred 24 hours after lead acetate injection and that such rats were no more sensitive to endotoxin than control rats at this time. While treatment

with methylprednisolone or cysteine reduced mortality in lead-acetate-sensitized rats exposed to endotoxin, it did not reverse hypophagocytosis of an R-E test-lipid emulsion (Cook and Di Luzio, 1973). Intravascular removal of an i.v. dose of S. enteritidis endotoxin was not altered by lead acetate injection (Filkins and Buchanan, 1973). Trejo and Di Luzio (1971) reported that colloidal carbon clearance by the R-E system was actually increased in lead-treated mice.

Treatment of rats with various sulfhydryl-containing compounds (cysteine, glutathione, methionine, and ethionine) reduced lead-induced sensitization to endotoxin (Bertok, 1968b). It was suggested that lead inactivation of sulfhydryl-containing enzymes or other compounds which play a part in endotoxin detoxification might be the mechanism of lead-induced sensitivity to endotoxin. Liver and spleen homogenates from rats which received i.v. injections of lead acetate had significantly lowered endotoxin detoxifying capacities (Trejo and Di Luzio, 1971; Trejo et al., 1972). Filkins and Buchanan (1973), however, found that liver homogenates from rats treated with lead acetate had no decreases in endotoxin-detoxifying potency and that the in vitro addition of lead to such homogenates did not alter detoxification.

Several workers have contended that altered hepatocyte function plays a more important role than does reduced

endotoxin detoxification in lead-induced endotoxin hypersensitivity (Cook and Di Luzio, 1973; Holper et al., 1973; Cook et al., 1974). A single i.v. injection of lead acetate caused ultrastructural alteration of both mitochondria and endoplasmic reticulum and accumulation of finely granular, electron-dense material in vesicles in hepatocytes of rats (Hoffman et al., 1972). Cook et al. (1973) observed more severe and diffuse ultrastructural changes in hepatocytes of rats exposed to lead acetate and endotoxin simultaneously than in rats receiving either of these individually.

Lead acetate-exposed rats had decreased bromsulfothalein (BSP) clearance, increased plasma concentrations of unconjugated bilirubin, and increased serum activities of alkaline phosphatase (SAP) and glutamic oxaloacetic transaminase (SGOT) (Trejo et al., 1972). Rats receiving lead acetate and endotoxin simultaneously had decreased BSP clearance, increased blood lactate and pyruvate concentrations, decreased plasma glucose concentrations, and increased serum glutamic pyruvic transaminase (SGPT), SGOT, and SAP activities (Cook and Di Luzio, 1973; Cook et al., 1973; Cook et al., 1974). These changes were partially prevented by methylprednisolone treatment.

Filkins (1973) observed that rats which received lead acetate and endotoxin experienced decreased gluconeogenesis from lactate, pyruvate, and alanine and had drastic depletion

of liver glycogen. It was felt that these findings indicated that lead-induced endotoxin hypersensitivity probably occurred due to a failure by the liver to make necessary metabolic adjustments to endotoxin and that one or more of the following alterations may have caused this lack of adjustment:

1. Decreased mitochondrial function.
2. Increased sensitivity of hepatocytes to insulin.
3. Decreased activity of an enzyme of the gluconeogenic pathway.

Rippe and Berry (1972) have noted that phosphoenolpyruvate carboxykinase, the rate limiting enzyme in gluconeogenesis, is inhibited in lead-treated mice.

The lipid soluble anti-oxidant, N,N'-diphenyl-p-phenylenediamine (DPPD) protects against lead-induced endotoxin hypersensitivity, and it is possible that an oxidizing action of lead plays a part in hepatocyte injury and increased endotoxin susceptibility (Trejo et al., 1972).

Trejo and Di Luzio (1971) suggested that lead absorption might lead to lysosomal instability with subsequent leakage of lysosomal enzymes and damage to Kupffer cells and hepatocytes. Increased serum activities of the lysosomal enzymes, beta-glucuronidase and acid phosphatase, have been detected in animals exposed to lead and endotoxin; however, since methylprednisolone treatment did not decrease serum activities of these enzymes, it is doubtful that lysosomal

instability is an important locus of lead-induced hypersensitivity to endotoxin (Cook and Di Luzio, 1973).

# MATERIALS AND METHODS

## Experimental Design

### Experiment 1

Thirty crossbred six-week-old pigs obtained from the Swine Nutrition Farm, Iowa State University and weighing 17-24 kilograms (kg) were randomly divided into 10 groups of 3 pigs per group. The pigs were housed in 10 pens in a heated barn. Pens had cement floors and bedding was not used.

All pigs were fed a balanced 16% protein ration which was replaced by a 14% protein ration as their weights increased. Feed consumption was limited in order to keep weight gains under 1 pound per day.

Each group was assigned 1 of 10 different treatments as shown in Table 1. The 3 pigs in group 5 received 3 different dosages of sodium acetate orally corresponding in acetate dosages to those received by groups 2, 3, and 4, respectively; and the 3 pigs in group 10 received 3 different dosages of sodium acetate intraperitoneally (i.p.) corresponding in acetate dosages to those received by groups 7, 8, and 9, respectively. Treatments were administered 6 days per week for 13 weeks (90 days).

Lead and sodium acetate were dissolved in distilled water in order to facilitate both oral and i.p. administration. Four different solutions of lead acetate were administered to

orally-exposed pigs. During the first 28 days of exposure, groups 1 and 2 received a solution containing 3.65 g/dl lead acetate and groups 3 and 4 received a solution containing

Table 1. Group treatments for experiment 1.

| Group | Compound | Route | Dosage | Pb Dosage |
|---|---|---|---|---|
| 1 | Pb Acetate | Oral | 8 mg/kg | 4.4 mg/kg |
| 2 | Pb Acetate | Oral | 16 mg/kg | 8.8 mg/kg |
| 3 | Pb Acetate | Oral | 32 mg/kg | 17.6 mg/kg |
| 4 | Pb Acetate | Oral | 64 mg/kg | 35.2 mg/kg |
| 5 | Na Acetate | Oral | --[a] | --- |
| 6 | Pb Acetate | i.p.[b] | 2 mg/kg | 1.1 mg/kg |
| 7 | Pb Acetate | i.p. | 4 mg/kg | 2.2 mg/kg |
| 8 | Pb Acetate | i.p. | 8 mg/kg | 4.4 mg/kg |
| 9 | Pb Acetate | i.p. | 16 mg/kg | 8.8 mg/kg |
| 10 | Na Acetate | i.p. | --[a] | --- |

[a]Variable dosages--see text.

[b]Intraperitoneal.

14.50 g/dl lead acetate. After 28 days, the concentrations of the solutions were increased in order to maintain a reasonable volume for administration, and groups 1 and 2 received a solution containing 5.45 g/dl lead acetate while groups 3 and 4 received a solution containing 21.80 g/dl lead

acetate. Oral sodium acetate solutions were also adjusted at 28 days in order to match the increased acetate dosages received by groups 2, 3, and 4. Sucrose was added to all oral solutions at a concentration of 2 g/dl in an attempt to increase palatibility.

Two different solutions were administered to i.p.-exposed pigs. During the first 28 days of exposure, groups 6 and 7 received a solution containing 1.82 g/dl lead acetate while groups 8 and 9 received a solution containing 7.30 g/dl lead acetate. Because of severe irritation of injection sites in pigs from groups 8 and 9, the pigs in these groups were given a solution identical to that administered to groups 6 and 7 after the 28th day of lead exposure.

Oral solutions were administered via disposable 12 milliliter (ml) polypropylene syringes with hubs inserted into 6 inch lengths of flexible plastic tubing.[1] As the pigs were held in an upright position, the tubing was thrust into the ventral-caudal oral cavity, and the solution was expelled. It was found that slow administration of the liquid stimulated the pig to swallow, and most of the solution was consumed.

Intraperitoneal solutions were administered via 12 ml polypropylene syringes with attached disposable 1 inch 20 gauge needles. Initially all injections were made through

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[1]Tygon tubing. The Norton Company, Akron, Ohio.

the ventral abdominal wall at a site caudal to the umbilicus and on either side of the midline. Later in the experiment, injections were made in the paralumbar fossa.

All pigs were weighed at weekly intervals. All groups were observed several times per day, and clinical signs were recorded. Body temperatures were measured when warranted by clinical signs. Blood samples were collected from all pigs via the orbital sinus (Huhn et al., 1969) prior to the beginning of lead or sodium acetate exposure and at weekly intervals thereafter. Blood was collected into tubes containing potassium ethylenediaminetetraacetate ($K_3$ EDTA), sodium heparin, or no anticoagulant.

Analyses on blood anticoagulated with $K_3$ EDTA were:

1. Hematocrit
2. Hemoglobin concentration
3. Total red blood cell count
4. Red blood cell morphology
5. Total leukocyte count
6. Leukocyte differential
7. Plasma protein concentration
8. Plasma fibrinogen concentration

Mean corpuscular volumes (MCVs), mean corpuscular hemoglobin concentrations (MCHCs), mean corpuscular hemoglobins (MCHs), absolute differential leukocyte counts, and plasma protein: fibrinogen ratios were calculated from these results.

Blood anticoagulated with sodium heparin was used in measuring lead concentration and delta-aminolevulinic acid dehydratase (ALAD) activity.

Serum was used in performing protein electrophoresis.

An erythrocyte osmotic fragility test was carried out during the 13th week of the experiment. Blood samples from 6 lead acetate-exposed and 6 sodium acetate-exposed pigs were tested. Potassium EDTA was the anticoagulant used in this trial.

Pigs which did not die during the 13 week lead or sodium acetate-exposure period were stunned by electric shock and exsanquinated. With the exception of groups 8 and 9, 1 of the 3 pigs from each group was euthanatized 1 day, 30 days, and 60 days, respectively, after the cessation of lead exposure. The 2 surviving pigs from groups 8 and 9 were euthanatized 1 day and 60 days, respectively, after the cessation of lead exposure.

Necropsy examinations were performed on all pigs, and all major organs were examined for macroscopic lesions. The following tissues were fixed in 10% formaldehyde solution in preparation for histologic examination:

1. Kidney
2. Liver
3. Cerebrum
4. Cerebellum

5. Spinal cord
6. Ischiatic nerve
7. Lymph node
8. Spleen
9. Lung
10. Other tissues with macroscopic lesions

Tissues collected, frozen, and later used for measurement of tissue lead concentrations were:

1. Kidney
2. Liver
3. Brain
4. Lymph node
5. Spleen
6. Lung
7. Skeletal muscle
8. Rib
9. Bone marrow

Experiment 2

Thirty-six eight- to nine-week-old crossbred pigs weighing 15-24 kg were obtained from the Swine Nutrition Farm, Iowa State University. The pigs were randomly divided into 2 groups (A and B) of 18 pigs per group and were housed in adjacent pens in a heated barn. The pens had cement floors, and no bedding was used.

All pigs were fed a balanced 16% protein ration. The ration received by group A also contained 500 parts per million (ppm) lead as lead acetate while that received by group B contained sodium acetate at a level such that the

acetate concentration was equivalent to that received by group A. It was estimated that lead-exposed pigs received a dosage of about 40 mg lead per kg body weight. This dosage was comparable to the highest oral lead exposure level in experiment 1. Rations were fed ad libitum.

At the beginning of the second week of lead or sodium acetate exposure and at weekly intervals thereafter, 6 pigs from each group were randomly selected, moved to an isolation building, and challenged with a live, virulent culture of Salmonella choleraesuis var. Kunzendorf[1] by i.p. injection. This resulted in the 1st replicate (1) being exposed to lead or sodium acetate for 1 week prior to challenge; the second replicate (2) being exposed for 2 weeks prior to challenge; and the 3rd replicate (3) being exposed for 3 weeks prior to challenge. Administration of lead or sodium acetate in the feed stopped following challenge.

Replicate 1 was challenged with 2 different dosages of a previously lyophilized and subsequently reconstituted culture of S. choleraesuis var. Kunzendorf which was incubated at 37°C for 2.5 hours after reconstitution. Three lead-exposed and 3 non-lead-exposed pigs received 2.5 ml (approximately $1.9 \times 10^9$ cells) while 3 lead-exposed and 3 non-lead-exposed

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[1]Courtesy of Dr. Billie O. Blackburn, Leader, Enteric Bacteriology, VSDL, APHIS, Ames, Iowa, 50010. NADC Type Culture #70-13679.

pigs received 0.5 ml (approximately $3.7 \times 10^8$ cells) of the culture.

Groups 2 and 3 were challenged with 5.0 ml ($>1.9 \times 10^9$ cells) of the same type of culture which had been incubated for 4 hours after reconstitution.

During lead or sodium acetate exposure and following Salmonella challenge, all pigs were observed 3-4 times per day, and clinical signs and mortalities were recorded. Body temperatures were measured 3 times per week during the lead or sodium acetate-exposure period, at 8 hours following Salmonella challenge, and at daily intervals thereafter.

Blood samples were collected from all pigs at various intervals via the orbital sinus (Huhn et al., 1969). Blood was collected into tubes containing sodium heparin, $K_3$ EDTA, or no anticoagulant.

Blood anticoagulated with sodium heparin was used for measuring lead concentration and ALAD activity.

Analyses on blood anticoagulated with $K_3$ EDTA were:

1. Hematocrit
2. Red blood cell morphology
3. Total leukocyte count
4. Leukocyte differential
5. Plasma protein concentration
6. Plasma fibrinogen concentration

Absolute differential leukocyte counts and plasma protein: fibrinogen ratios were calculated from these results.

Serum was used in performing protein electrophoresis, measuring complement titers, and determining S. choleraesuis var. Kunzendorf agglutinin titers.

A schedule of analyses is given in Table 2.

Pigs which did not die during the experiment were stunned by electric shock and exsanquinated 10 days after challenge. Necropsy examinations were performed on all pigs, and all major organs were examined for macroscopic lesions. The following tissues were fixed in 10% formaldehyde solution in preparation for histologic examination:

1. Liver
2. Kidney
3. Mesenteric lymph node
4. Spleen
5. Lung
6. Ileum
7. Colon
8. Cecum

Mesenteric lymph node, mandibular lymph node, liver, kidney, spleen, and ileum were collected and cultured immediately.

Portions of kidney, liver, lymph node, and brain were frozen and later analyzed for lead concentrations.

Table 2. Schedule of analyses on blood samples collected during experiment 2.

| Day | Analyses[a] | Replicates Tested |
|---|---|---|
| Exposure Period[b] | | |
| 0 | Pb, ALAD, CBC, Elect, C' | 1,2,3 |
| 2 | Pb, ALAD, Elect, C' | 1,2,3 |
| 4 | Pb, ALAD, Elect, C' | 1,2,3 |
| 8 | Pb, ALAD, CBC, Elect, C' | 1,2,3 |
| 15 | Pb, ALAD, CBC, Elect, C' | 2,3 |
| 21 | Pb, ALAD, CBC, Elect, C' | 3 |
| Post-challenge | | |
| 2 | Pb, ALAD, CBC, Elect, C', Agglut | 1,2,3 |
| 4 | CBC, Elect, C', Agglut | 1,2,3 |
| 7 | Elect, C', Agglut | 1,2,3 |
| 9 | Pb, ALAD, CBC, Elect, C', Agglut | 1,2,3 |

[a]Pb = Blood lead concentration; ALAD = Blood delta-aminolevulinic acid dehydratase activity; CBC = Hematocrit, total leukocyte count, leukocyte differential, plasma protein and fibrinogen concentrations; Elect = Serum protein electrophoresis; C' = Serum complement titration; Agglut = Serum S. choleraesuis var. Kunzendorf agglutinin titers.

[b]Period of lead or sodium acetate exposure prior to Salmonella challenge.

Experiment 3

Twenty-four four- to five-week-old crossbred pigs derived from breeding stock at the Swine Nutrition Farm, Iowa State University, and weighing 5-9 kg were randomly divided into 3 groups (A, B, and C) of 8 pigs each. Each group was divided into 2 replicates of 4 pigs each. The animals were housed in an isolation building with each replicate in a separate pen. Pens had cement floors, and no bedding was used. Because the pigs were young and the weather was cold, a heat lamp was provided for each pen.

Lead-exposed pigs were fed a balanced 16% protein ration containing 3,000 ppm lead as lead chloride. This resulted in an estimated lead exposure of 250 mg lead per kg body weight. Control pigs were fed the 16% protein ration containing sodium chloride rather than lead chloride. The chloride concentrations of both the lead chloride- and sodium chloride-containing rations were equivalent. All pigs were fed ad libitum. The groups were designated as follows: Group A--not exposed to lead; Group B--exposed to lead for 2 weeks prior to challenge with S. choleraesuis var. Kunzendorf; and Group C--exposed to lead for 1 week prior to challenge with S. choleraesuis var. Kunzendorf.

All pigs were challenged simultaneously with a virulent strain of S. choleraesuis var. Kunzendorf.[1] Twelve ml of a bacterial suspension (see laboratory procedures) was administered intragastrically via a flexible plastic stomach tube[2] to each of the 24 pigs. After the tube was inserted into the stomach, a 20 ml polypropylene syringe containing the bacterial suspension was attached to the free end, and the suspension was expelled. The tube was flushed with 20 ml of sterile saline to insure complete administration of the suspension. At the time of challenge, lead and sodium chloride exposure was terminated.

Rectal swabs and serum samples were collected from all pigs prior to their assignment to isolation pens. Swabs were cultured (see laboratory procedures), and serum samples were tested for S. choleraesuis var. Kunzendorf agglutinin titers.

Throughout the trial, all pigs were observed at least 4 times per day, and clinical signs and mortalities were recorded. In an attempt to quantitate the clinical signs observed, a numerical rating system was used (Table 3) following Salmonella challenge. Body temperatures of all pigs were measured and recorded daily beginning 1 week prior to lead or sodium chloride exposure.

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[1] Courtesy of R. D. Glock, DVM, Department of Veterinary Pathogy, Iowa State University, Ames, Iowa.

[2] Tygon tubing. The Norton Company, Akron, Ohio.

Table 3. Numerical rating system for clinical signs--Experiment 3.

| Rating | General condition | Fecal consistency | Appetite | Hydration |
|---|---|---|---|---|
| 1 | Normal | Normal | Normal | Normal |
| 2 | Rises readily but depressed. Slightly gaunt. | Soft(pasty) | Slightly anorectic. | Skin slightly less pliable. |
| 3 | Rises reluctantly. Very gaunt. | Loose(liquid) | Interested in feed, but eats little. | Skin not pliable. Eyes slightly sunken. |
| 4 | Moribund (cannot rise). | Watery | Completely anorectic. | Skin not pliable. Eyes markedly sunken. |

Immediately preceding initiation of lead or sodium chloride exposure and at pre-determined intervals throughout the experiment, blood was collected from the anterior vena cava (Hoerlein et al., 1951) into tubes containing sodium heparin, $K_3$ EDTA, or no anticoagulant.

Blood anticoagulated with sodium heparin was used in measuring lead concentration and ALAD activity.

Blood anticoagulated with $K_3$ EDTA was used in the following determinations:

1. Hematocrit
2. Hemoglobin concentration
3. Total red blood cell count
4. Red blood cell morphology
5. Total leukocyte count
6. Leukocyte differential
7. Plasma protein concentration
8. Plasma fibrinogen concentration

Mean corpuscle volumes, MCHCs, MCHs, absolute leukocyte differentials, and plasma protein:fibrinogen ratios were calculated from these results.

Serum was used in performing protein electrophoresis and in determining S. choleraesuis var. Kunzendorf agglutinin titers.

A schedule of analyses on blood samples is given in Table 4.

All pigs which had not died by 14 days after challenge were stunned by electric shock and exsanquinated. Necropsy examinations were performed, and all major organs were examined for macroscopic lesions. The following tissues were fixed in 10% formaldehyde solution in preparation for histologic examination:

1. Liver
2. Kidney
3. Spleen

Table 4. Schedule of analyses on blood samples collected during experiment 3.

| Day | Analysis[a] |
|---|---|
| 1 | Pb[b], ALAD[b], CBC |
| 6 | Pb, ALAD, CBC, Elect |
| 8 | Pb, ALAD, CBC |
| 13 | Pb, ALAD, CBC, Elect, Agglut |
| 15[c] | Pb, ALAD, CBC |
| 17 | CBC, Elect, Agglut |
| 18 | CBC |
| 20 | CBC, Elect, Agglut |
| 21 | Pb, ALAD |
| 22 | CBC, Elect, Agglut |
| 24 | CBC, Agglut |

[a]Pb = Blood lead concentration; ALAD = Blood delta-aminolevulinic acid dehydratase activity; CBC = Hematocrit, blood hemoglobin concentration, total red blood cell count, total leukocyte count, leukocyte differential, plasma protein and fibrinogen concentrations; Elect = Serum protein electrophoresis; Agglut = S. choleraesuis var. Kunzendorf agglutinin titers.

[b]No blood lead concentrations or ALAD activities measured in group C on this day.

[c]Day of challenge.

4. Mesenteric lymph node
5. Lung
6. Duodenum
7. Jejunum
8. Ileum
9. Colon
10. Cecum

Mesenteric lymph node, liver, and ileum were collected and cultured immediately.

Portions of liver and kidney were frozen and later analyzed for lead concentrations.

## Laboratory Procedures

### Preparation of inocula

Experiment 2 *Salmonella choleraesuis* var. *Kunzendorf* organisms were transferred from an agar slant to 4 ml of trypticase soy broth and incubated at 37°C for 8 hours. The 4 ml culture was then added to 100 ml of trypticase soy broth and incubated at 37°C for 6 hours. At the end of this time, 12.5 ml of the culture was transferred into 250 ml of trypticase soy broth and incubated at 37°C for 4 hours. Ten ml aliquots of the final culture were pipetted into 15 ml serum bottles and lyophilized in a Universal Sub-Mobile-15 lyophilizer.[1] The bottles were then capped with rubber

[1]The Virtis Company, Gardiner, New York.

stoppers and aluminum seals. Prior to challenge, the lyophilized cultures were reconstituted with 10 ml of distilled water and incubated in a 37°C waterbath.

Experiment 3 Standard 0.1 ml aliquots from a frozen culture of S. choleraesuis var. Kunzendorf were placed in 4 ml of trypticase soy broth and incubated for 24 hours at 37°C. After incubation, the 4 ml quantities were transferred into 250 ml of fresh trypticase soy broth in 500 ml bottles. These were incubated for 5 hours at 37°C with continuous shaking on a variable speed rotator. The cultures were then transferred to 50 ml sterile, screw-capped centrifuge bottles, and the bacterial cells were sedimented by centrifugation at 1400 x g for 30 minutes. The packed cells were washed 3 times by alternate resuspension and centrifugation in saline. After washing, the bacterial cells were diluted in saline to a density equal to tube 7 of a McFarland nephalometer (approximately 2 x $10^9$ bacteria/ml).

## Blood lead concentrations

Blood lead concentrations were measured on an atomic absorption spectrophotometer.[1] The method of extraction and measurement was that described by Hessel (1968).

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[1]Perkin-Elmer Model 303 Atomic Absorption Spectrophotometer. Perkin-Elmer, Inc., Maywood, Illinois.

Blood ALAD activities

The method of Burch and Siegel (1971) was used to assay blood ALAD activities. Erythrocytes in whole blood were lysed by the addition of a 0.2% solution of Triton X-100[1], and delta-aminolevulinic acid (ALA) hydrochloride[1] was added to the hemolysate. During a 1 hour incubation period at 38°C, ALAD from hemolyzed erythrocytes catalyzed the condensation of ALA to form porphobilinogen. The addition of trichloroacetic acid to the hemolysate-enzyme mixture precipitated proteins and stopped the reaction. The mixture was centrifuged, and the supernatant fluid was decanted into other tubes. Addition of a modified Erhlich's reagent to the supernatant fluid caused a color to develop. The absorbance of the solution was measured on a Coleman Junior II Spectrophotometer[2] at 555 nanometers (nm). A unit of enzyme activity was expressed as an increase in absorbance of 0.100 with a 1.0 cm light path per ml of erythrocytes per hour at 38°C.

Hematologic measurements

Hematocrits were measured by the microhematocrit method. The cyanmethemoglobin method[3] was used in determining blood

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[1]Sigma Chemical Company, St. Louis, Missouri.

[2]Perkin-Elmer, Inc., Maywood, Illinois.

[3]Hycel, Inc., Houston, Texas.

hemoglobin concentrations. Total red blood cell and leukocyte counts were performed on a Coulter Counter Model F[1] electronic cell counter. Mean corpuscular volumes, MCHCs, and MCHs were calculated via the standard Wintrobe formulas (Schalm et al., 1975). Blood smears were made within 3 hours of collection, fixed with absolute methanol, and stained with Wright's stain (Schalm et al., 1975). The cross-sectional method (Schalm et al., 1975) was used in performing differential leukocyte counts. Red blood cell morphology was observed during differential counting. Plasma and serum protein concentrations were measured by refractometry.[2] Plasma fibrinogen concentrations were determined via the heat precipitation method (Schalm et al., 1975).

Erythrocyte fragility test

The method used in measuring erythrocyte fragility was that of Hepler (1966). One drop of whole blood was added to each of 20 tubes containing dilutions of sodium chloride ranging from 0.28%-0.66%. Each tube contained a dilution 0.02% more concentrated than the previous tube. The mixtures were incubated at room temperature for 2 hours. Dilutions at which initial hemolysis (faint yellow color in supernatant fluid) and complete hemolysis (red color in supernatant

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[1]Coulter Electronics, Inc., Hialeah, Florida.

[2]American Optical TS Meter. American Optical Instruments Company, Buffalo, New York.

fluid and no erythrocytes on bottom of tube) occurred were recorded.

Serum agglutinin titrations

Production of antigens for serum agglutinin titrations was carried out by propagating cultures of S. choleraesuis var. Kunzendorf in Kolle flasks containing tryptose agar. After incubation for 24 hours, the organisms were suspended in sterile saline, diluted with 3 parts 95% ethyl alcohol, and incubated at 37°C for 2 hours. Following centrifugation and elimination of the supernatant fluid, the bacterial cells were diluted in phenolized saline (0.3%) to a concentration equal to tube 3 of a McFarland nephalometer. The resulting antigen suspension was somatic in type.

Flagellar antigens were produced in a similar manner except for the elimination of ethyl alcohol dilution and incubation, and the replacement of phenolized saline with formalinized saline (0.5%).

Preliminary agglutinin titers were determined by diluting serum samples 1:5, 1:25, 1:50, 1:100, and 1:200 in phosphate-buffered saline and adding a preparation of flagellar antigen. Test results were determined after 2 hours incubation at 37°C and after 18 hours incubation at 55°C. Proper negative controls were used.

Post-challenge agglutinin titrations were performed by making two-fold dilutions of test sera in saline. Dilutions

ranged from 1:10 to 1:2560. Saline was used as a negative control. After addition of somatic or flagellar antigen preparations (resulted in dilutions becoming 1:20 through 1:5120), the tubes were incubated at 52°C. Flagellar agglutinin titers were read after 4 hours of 52°C and 18 hours of room temperature incubation.

Positive flagellar agglutination tests were indicated by a cottony, flocculent mass of bacteria floating in the mixture. A mat or button of bacteria which was not readily dispersed by shaking represented a positive somatic agglutination reaction.

Serum protein electrophoresis

Experiment 1 Serum protein electrophoresis was performed in a Mikrophor electrophoresis chamber[1] using cellulose acetate membranes[2] and barbital-sodium barbital-sodium acetate buffer[3] (pH 8.6 and molarity of 0.05). Serums were electrophoresed for 50 minutes at a constant voltage of 250 volts. After electrophorectic migration, protein bands were stained with Ponceau S.[4]

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[1]Brinkman-Sartorius, Brinkman Instruments, Westbury, New York.

[2]Brinkman Instruments, Westbury, New York.

[3]Kallestad Labs, Inc., Minneapolis, Minnesota.

[4]Gelman Instrument Company, Ann Arbor, Michigan.

Experiments 2 and 3 Serum protein electrophoresis was performed in an ACI Agarose Universal Electrophoresis Cell[1] using ACI Agarose Universal Electrophoresis Film[1] and Universal Barbital Buffer[1] (pH 8.6, molarity of 0.05, containing 0.035% EDTA). Serums were electrophoresed for 35 minutes at a constant voltage of 110 volts. After electrophoretic migration, protein bands were stained with Amido Black 10B.[2]

Relative serum protein concentrations were quantitated with a scanning photoelectric densitometer.[3] Optimal density was measured at 600 nm (experiment 1) or 580 nm (experiments 2 and 3), and an integrated density curve was plotted. Individual peaks were separated by drawing vertical lines at their lowest points, and the number of integrated pulse strokes falling under each peak were counted and used in calculating relative percentages of each fraction. Relative percentages were converted to absolute concentrations by multiplying serum protein concentrations by the percentage of each protein fraction.

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[1]Analytical Chemists, Inc., Palo Alto, California.

[2]Gelman Instrument Company, Ann Arbor, Michigan.

[3]Model RIIV, Beckman Instruments, Fullerton, California.

Serum complement titration

Equal volumes of a sheep red blood cell suspension ($4 \times 10^8$ cells/ml) which had previously been sensitized by incubation with 8 $AbH_{50}$ units (1 $AbH_{50}$ unit represents a quantity of hemolysin which will lyse 50% of the erythrocytes in the presence of adequate complement) of hemolytic antiserum were added to serial dilutions of test serum in a barbital buffer. Cell suspensions were also added to control tubes containing diluent but no serum. After incubation for 1 hour at 37°C, the mixtures were centrifuged in the cold at 1000 x g for 5 minutes (in order to sediment unlysed erythrocytes). Supernatant fluid was decanted into spectrophotometer cells and optical density (O.D.) was read at 530 nm. Transmission was considered to be 100% in a diluent blank, and the O.D. of a completely hemolyzed sample which contained the same concentration of erythrocytes as test mixtures was also measured. Percent lysis was calculated via the formula:

$$\% \text{ lysis} = \frac{\text{O.D. test serum} - \text{O.D. control}}{\text{O.D. 100\% hemolyzed} - \text{O.D. control}} \times 100$$

The complement titer was the dilution at which 50% lysis took place.

Salmonella isolation

Tissues and swabs to be cultured for Salmonella were placed in tetrathionate broth and incubated for 18 hours at 37°C. Streakings were made from tetrathionate broth to

brilliant green agar, incubated at 37°C for 24 hours, and examined for the presence of lactose negative (red) Salmonella colonies. Selected isolates were transferred to tryptose broth, incubated at 37°C for 24 hours, and serotyped.[1]

Tissue lead concentrations

Tissues were dried in an oven overnight, and then ashed in a muffle furnace at 450°C. Lead was extracted from the ash into 2N hydrochloric acid and aspirated into an atomic absorption spectrophotometer.[2] The tissue concentration of lead was calculated using a standard curve.

Tissue preparation for histologic examination

Formalin-fixed tissues were trimmed and prepared for sectioning by standard ethanol dehydration and paraffin embedding techniques. Sections cut 6 microns thick were stained with Harris hematoxylin and eosin Y. In addition, sections of liver and kidney were stained with Ziehl-Neelsen acid-fast stain (Mallory, 1942).

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[1]Courtesy of Dr. Billie O. Blackburn, Leader, Enteric Bacteriology, VSDL, APHIS, Ames, Iowa.

[2]Perkin-Elmer Model 303 Atomic Absorption Spectrophotometer, Perkin-Elmer, Inc., Maywood, Illinois.

## Statistical Procedures

### Experiment 1

Since lead or sodium acetate solutions were administered to pigs on an individual basis, each pig within a group was considered an experimental unit. This resulted in 5 orally- and 5 i.p.-exposed groups with 3 replicates per group.

An unpaired group comparison was used to analyze for group differences (lead-treated versus control) in erythrocyte fragility. The standard error of the difference between group means was calculated via standard formulas (Steel and Torrie, 1960). A Student's t test was applied to evaluate the significance of this difference.

In evaluating other results, orally- and i.p.-exposed groups were analyzed separately. The experimental design was considered to be a split-plot. An analysis of variance was performed with group and replicate within group considered the whole-plot portion and week, group by week interaction, and residual error constituting the split-plot portion. The group means for each parameter were calculated by averaging the 13 values obtained following initiation of lead or sodium acetate exposure. Standard errors of these means and least significant differences were calculated using appropriate formulas (Cochran and Cox, 1957). For any given parameter, means of groups exposed to lead orally were

compared to that of the orally sodium acetate-exposed group, and means of groups exposed to lead i.p. were compared to that of the i.p. sodium acetate-exposed group.

Experiment 2

Each of the replicates challenged after 1 week, 2 weeks, or 3 weeks of lead or sodium acetate exposure was considered an experimental unit. Thus, there were 2 groups, lead acetate-exposed and sodium acetate-control, and 3 replicates within each of these groups.

The experimental design was considered to be a split-plot. In analyzing blood lead concentrations and ALAD activities, group and replicate within group were considered the whole-plot portion, and day, group by day interaction, and residual error constituted the split-plot portion of the analysis.

For analysis of all parameters other than blood lead concentrations, blood ALAD activities and serum agglutinin titers, group and replicate within group were considered the whole-plot portion and period (period 1 = pre-challenge and period 2 = post-challenge), day within period, group by period interaction, group by day within period interaction, and residual error constituted the split-plot portion of the analysis.

Standard errors of means and least significant differences were calculated by standard formulas (Cochran and Cox, 1957).

In testing for significant differences in blood lead concentrations and ALAD activities, group means were calculated by averaging values obtained from the 3 replicates for the entire experiment. Two means resulted, 1 for the lead acetate-exposed group and 1 for the sodium acetate-exposed control group.

In testing for significant differences of parameters other than blood lead concentrations, blood ALAD activities, and serum agglutinin titers, means were calculated for the lead acetate-exposed group and the sodium acetate-exposed control group within each period, i.e. 4 means were obtained: lead acetate-exposed prior to challenge, lead acetate-exposed following challenge, sodium acetate-control prior to challenge, and sodium acetate-control following challenge. The 2 means prior to challenge were compared, and the 2 means following challenge were compared.

Serum agglutinin titers were compared on a daily basis. An unpaired group comparison was performed using standard formulas (Steel and Torrie, 1960) to calculate the standard error of the difference between means of lead acetate-exposed and sodium acetate-control groups, and a Student's t test was used to evaluate significance.

Experiment 3

There were 3 groups and 2 replicates within each group. Each replicate was considered an experimental unit. Results were analyzed as in experiment 2 with the exception that means from 3 groups rather than 2 groups were analyzed. Period 1 was again considered to be pre-challenge, and period 2 was considered to be post-challenge.

# RESULTS

## Experiment 1

### Clinical signs and mortalities

None of the pigs exposed to lead acetate orally died during the experiment. Clinical abnormalities were observed in only 2 of these pigs; both were from group 4 (64 mg/kg lead acetate orally). One pig had a cough after 6 days of lead exposure, and this persisted for the remainder of the experiment. After 8 days of lead exposure, both pigs were gaunt, had rough coats, and had body temperatures of 40°C. These abnormalities lasted for 5 to 6 days.

One pig from group 5 (oral sodium acetate-control) began coughing sporadically after 6 days of sodium acetate exposure and continued to do so for the remainder of the experiment. No other clinical signs were seen in the oral-control group.

Four of the pigs exposed to lead acetate i.p. died during the experiment, and clinical signs were observed in all i.p. lead acetate-exposed groups. Signs ranged from gauntness and rough coats in those most mildly affected to depression, apparent abdominal discomfort, pale mucous membranes and skin, variable degrees of inappetance, and vomiting in pigs more severely ill. Initial signs appeared from 3 to 49 days after the beginning of lead exposure. Many of the pigs developed masses in the abdominal wall at injection sites. Such masses

were first noticed after 16 days of lead exposure. Pigs from groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.) were more severely affected than those from groups 6 (2 mg/kg lead acetate i.p.) and 7 (4 mg/kg lead acetate i.p.).

Two pigs from group 8 died after 25 and 35 days of lead exposure, respectively; and 2 pigs from group 9 died after 23 and 38 days of lead exposure, respectively. Pigs became incoordinated and reluctant to move several days before death. One pig had diarrhea and another had lateral nystagmus and rhythmic jerking of the head and forelimbs prior to death.

Pigs exposed to sodium acetate i.p. remained clinically normal throughout the experiment.

Blood lead concentrations

Highly significant ($P < .01$) elevations of mean blood lead concentrations for the entire experiment occurred in all lead acetate-exposed groups.

Prior to lead or sodium acetate exposure, blood lead concentrations were at background levels (mean = 6 ug/dl). Blood lead concentrations rose gradually throughout the experiment in all groups exposed to lead acetate orally and in i.p. lead acetate-exposed groups 6, 7, and 8. Blood lead concentrations rose much more rapidly in group 9 (16 mg/kg lead acetate i.p.), and the surviving pig in this group had

a blood lead concentration of 14,300 ug/dl by the end of the 13th week of lead exposure. The highest blood lead concentration detected in orally lead acetate-exposed pigs was 290 ug/dl at the end of the 13th week in a pig from group 4 (64 mg/kg lead acetate orally).

Highest blood lead concentrations in the 4 pigs which died before the end of the experiment were 60, 69, 93, and 122 ug/dl. Several other pigs had blood lead concentrations exceeding these levels during the experiment.

Blood lead concentrations of control pigs remained at background levels throughout the trial.

Mean blood lead concentrations and results of statistical analyses are given in Tables 5 and 6. Weekly group means are given in the Appendix (Tables 36 and 37).

Blood ALAD activities

Highly significant ($P < .01$) reductions of mean blood ALAD activities for the entire experiment were detected in all lead acetate-exposed groups.

Mean blood ALAD activity prior to initiation of lead exposure was 214.4 units. After 1 week of lead exposure, activities of lead acetate-exposed groups had decreased sharply to 20.2-30.8% of their original levels. These activities continued to fall for the first 8 weeks of the experiment and then became stable at very low levels.

Table 5. Experiment 1--Orally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 1 | 2 | 3 | 4 | 5 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Blood lead concentration (ug/dl) | 38** | 38** | 54** | 78** | 10 | 14 | 20 |
| Blood ALAD activity | 15.2** | 10.8** | 11.0** | 8.2** | 130.8 | 15.3 | 21.8 |
| Blood hemoglobin concentration (g/dl) | 12.2 | 12.5 | 12.3 | 11.7 | 12.5 | 0.9 | 1.3 |
| Hematocrit (%) | 38.0 | 38.8 | 38.2 | 37.1 | 37.1 | 2.8 | 4.0 |
| Total red blood cell count (x $10^6$)[c] | 6.90 | 7.03 | 6.70 | 6.96 | 7.00 | 0.62 | 0.89 |
| MCV (fl) | 56.0 | 56.4 | 57.8 | 54.1 | 56.8 | 3.8 | 8.3 |
| MCHC (g/dl) | 32.2 | 32.2 | 32.2 | 31.6 | 32.2 | 0.9 | 1.3 |
| MCH (pg) | 18.1 | 18.1 | 18.6 | 17.2 | 18.2 | 2.0 | 2.9 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 1, 2, 3, and 4 received 8, 16, 32, and 64 mg/kg lead acetate, respectively.

[c]Count in cells/cubic mm.

**Highly significant difference ($P < .01$) as compared to group 5.

Table 6. Experiment 1--Intraperitoneally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 6 | 7 | 8 | 9 | 10 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Blood lead concentration (ug/dl) | 64* | 82* | 89* | 1,940** | 12 | 50 | 81 |
| Blood ALAD activity | 12.8** | 13.5** | 12.4** | 11.1** | 80.6 | 29.2 | 42.3 |
| Blood hemoglobin concentration (g/dl) | 11.2 | 11.4 | 10.1* | 9.0** | 12.5 | 2.3 | 3.2 |
| Hematocrit (%) | 35.8 | 37.0 | 32.5* | 30.1* | 35.5 | 2.8 | 4.0 |
| Total red blood cell count (x $10^6$)[c] | 6.49 | 6.59 | 6.26 | 5.48 | 6.66 | 2.26 | 3.24 |
| MCV (fl) | 56.2 | 56.9 | 52.3* | 55.2 | 58.5 | 4.1 | 6.0 |
| MCHC (g/dl) | 31.2** | 30.8** | 31.2** | 29.9** | 32.6 | 0.5 | 0.8 |
| MCH (pg) | 17.5** | 17.5** | 16.2** | 16.4** | 19.0 | 1.3 | 2.0 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 6, 7, 8, and 9 received 2, 4, 8, and 16 mg/kg lead acetate, respectively.

[c]Count in cells/cubic mm.

*Significant difference ($P < .05$) as compared to group 10.

**Highly significant difference ($P < .01$) as compared to group 10.

Blood ALAD activities of sodium acetate-control groups declined gradually throughout the trial. By the end of the experiment, mean activities were 39 and 15% of their original levels in oral- and i.p.-control groups, respectively.

Mean blood ALAD activities and results of statistical analyses are given on Tables 5 and 6. Weekly group means are given in the Appendix (Tables 38 and 39).

Hematologic observations

There were significant ($P < .05$) and highly significant ($P < .01$) decreases in both mean blood hemoglobin concentrations and hematocrit percentages for the entire experiment in groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.), respectively. Weekly mean blood hemoglobin concentrations were below normal (10-16 g/dl) in groups 8 and 9 on most sampling dates after 5 weeks of lead exposure, and weekly mean hematocrit percentages were below normal (32-50%) in these same groups on most bleeding dates after 3 weeks of lead exposure. Hemoglobin concentrations and hematocrit percentages remained within normal limits in all other groups.

No significant group differences in mean total red blood cell counts for the entire trial were detected. The red blood cell count of the surviving pig from group 9 (16 mg/kg lead acetate i.p.) was below normal (5-8 x $10^6$/cubic mm) on several bleeding dates after 7 weeks of lead exposure.

A significant ($P < .05$) decrease in mean MCV for the entire experiment occurred only in group 8 (8 mg/kg lead acetate i.p.). Although weekly mean MCVs were occasionally below normal (50-68 fl) in all groups, low values were more consistently found in group 8.

Highly significant ($P < .01$) decreases in mean MCHCs and MCHs for the whole experiment were found in all i.p. lead acetate-exposed groups. Weekly mean MCHCs fell below normal (30-34 g/dl) only in i.p. lead acetate-exposed groups, and such low values occurred as early as 1 week after initiation of lead exposure. Weekly mean MCHs were below normal (17-21 pg) in all groups (both orally and i.p. lead and sodium acetate-exposed) on various sampling dates.

Mean blood hemoglobin concentrations, hematocrit percentages, total red blood cell counts, MCVs, MCHCs, and MCHs and results of statistical analyses are summarized in Tables 5 and 6. Weekly groups means are given in the Appendix (Tables 40-51).

Variable degrees of anisocytosis and polychromasia plus small numbers of nucleated red blood cells were evident in smears from both lead acetate-exposed and sodium acetate-control pigs during the trial. These changes were most marked in the surviving pigs from groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.). Basophilic stippling was occasionally observed in erythrocytes of both

orally and i.p. lead acetate-exposed pigs but not in those of sodium acetate-controls.

A highly significant ($P < .01$) increase in mean total leukocyte count for the whole trial occurred in group 9 (16 mg/kg lead acetate i.p.). Although weekly mean total leukocyte counts were occasionally above normal (11,000-22,000/cubic mm) in all groups, they were most consistently elevated in group 9 (on 10 of 14 bleedings).

A highly significant ($P < .01$) increase in mean total segmented neutrophil count for the entire trial was also detected in group 9, but a significant ($P < .05$) decrease in this same value occurred in group 3 (32 mg/kg lead acetate orally). Weekly mean total segmented neutrophil counts were above or below normal (3,200-10,000/cubic mm) in most groups at various times, but were most consistently and markedly elevated in group 9.

Highly significant ($P < .01$) increases in mean total band neutrophil counts for the entire experiment were noted in groups 2 (16 mg/kg lead acetate orally) and 9 (16 mg/kg lead acetate i.p.). Weekly mean total band neutrophil counts were above normal (0-800/cubic mm) on several bleeding dates in all groups. These counts were most consistently elevated in i.p. lead and sodium acetate-exposed groups, and highest counts occurred in group 9.

The mean total lymphocyte count for the entire trial was significantly ($P < .05$) decreased in group 8 (8 mg/kg lead acetate i.p.). Mean total lymphocyte counts were above normal (4,500-13,000/cubic mm) on certain sampling dates in groups 1 (8 mg/kg lead acetate orally), 4 (64 mg/kg lead acetate orally), 5 (oral-control), and 10 (i.p.-control) and below normal on the final bleeding of group 9 (16 mg/kg lead acetate i.p.).

The mean total eosinophil count for the whole trial was significantly ($P < .05$) elevated in group 7 (4 mg/kg lead acetate i.p.). Weekly mean total eosinophil counts were never above normal (50-2,000/cubic mm) in any group and were below normal in all groups on 1 or more bleeding dates. Weekly counts tended to be highest in group 7 and lowest in group 9 (16 mg/kg lead acetate i.p.).

There were no significant group differences in mean total monocyte or basophil counts for the entire experiment. While weekly mean total monocyte counts were below normal (50-2,000/cubic mm) on 1 or more bleeding dates in several orally and i.p. lead acetate-exposed groups, they were most consistently low in group 9 (16 mg/kg lead acetate i.p.). Weekly mean total basophil counts were within normal limits (0-400/cubic mm) except for a low value detected on the last sampling date in group 6 (2 mg/kg lead acetate i.p.).

The mean plasma protein:fibrinogen ratio for the entire trial was significantly ($P < .05$) decreased in group 9 (16 mg/kg lead acetate i.p.). Although weekly mean plasma protein:fibrinogen ratios fluctuated in all groups, they were most consistently low in group 9.

Mean total leukocyte, segmented neutrophil, band neutrophil, lymphocyte, eosinophil, monocyte, and basophil counts and mean plasma protein:fibrinogen ratios are summarized in Tables 7 and 8. Results of statistical analyses are also presented in these tables. Weekly group means are given in the Appendix (Tables 52-67).

Erythrocyte osmotic fragility

Initial hemolysis occurred at mean sodium chloride concentrations of 0.47% and 0.50% and complete hemolysis was observed at mean sodium chloride concentrations of 0.39% and 0.41% in lead acetate-exposed and sodium acetate-control pigs, respectively. In comparing lead acetate-exposed and sodium acetate-control pigs, differences in concentrations at which initial or complete hemolysis took place were not statistically significant.

Cellulose acetate electrophoresis

A significant ($P < .05$) decrease of mean serum protein concentration for the whole trial was found only in group 3 (32 mg/kg lead acetate orally). Weekly mean total serum

Table 7. Experiment 1--Orally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 1 | 2 | 3 | 4 | 5 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Total counts[c] | | | | | | | |
| Leukocyte | 19,684 | 17,931 | 18,231 | 20,762 | 18,315 | 4,048 | 5,758 |
| Segmented neutrophil | 5,203 | 5,633 | 4,397* | 5,922 | 6,548 | 1,916 | 2,726 |
| Band neutrophil | 693 | 1,906** | 748 | 864 | 932 | 674 | 957 |
| Lymphocyte | 12,919 | 9,093 | 11,945 | 12,607 | 10,003 | 3,859 | 5,489 |
| Eosinophil | 455 | 616 | 660 | 723 | 390 | 417 | 593 |
| Monocyte | 343 | 432 | 379 | 540 | 390 | 243 | 346 |
| Basophil | 95 | 81 | 80 | 86 | 52 | 51 | 73 |
| Plasma protein: fibrinogen ratio | 34 | 36 | 30 | 36 | 31 | 8 | 11 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 1, 2, 3, and 4 received 8, 16, 32, and 64 mg/kg lead acetate, respectively.

[c]Cells/cubic mm of blood.

*Significant difference ($P < .05$) as compared to group 5.

**Highly significant difference ($P < .01$) as compared to group 5.

Table 8. Experiment 1--Intraperitoneally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 6 | 7 | 8 | 9 | 10 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Total counts[c] | | | | | | | |
| Leukocyte | 19,109 | 20,969 | 18,501 | 25,733** | 20,231 | 3,002 | 4,414 |
| Segmented neutrophil | 5,287 | 7,013* | 6,529 | 10,189** | 5,634 | 1,358 | 2,006 |
| Band neutrophil | 1,057 | 1,554 | 1,672 | 4,892** | 1,897 | 924 | 1,364 |
| Lymphocyte | 11,480 | 10,722 | 9,205* | 9,889 | 11,838 | 2,219 | 3,252 |
| Eosinophil | 725 | 1,059* | 440 | 292 | 616 | 364 | 534 |
| Monocyte | 391 | 481 | 542 | 369 | 412 | 339 | 501 |
| Basophil | 145 | 135 | 63 | 38 | 104 | 54 | 81 |
| Plasma protein: fibrinogen ratio | 30 | 37 | 42 | 24* | 36 | 10 | 15 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 6, 7, 8, and 9 received 2, 4, 8, and 16 mg/kg lead acetate, respectively.

[c]Cells/cubic mm of blood.

*Significant difference ($P < .05$) as compared to group 10.

**Highly significant difference ($P < .01$) as compared to group 10.

protein concentrations of group 3 were lower than those of other orally lead or sodium acetate-exposed groups on most sampling dates.

Highly significant ($P < .01$) decreases of mean serum albumin concentrations for the entire experiment were detected in groups 3 (32 mg/kg lead acetate orally), 4 (64 mg/kg lead acetate orally) and all i.p. lead acetate-exposed groups. After 2 weeks of lead exposure, weekly mean serum albumin concentrations in these orally or i.p. lead acetate-exposed groups were lower than those of oral- or i.p.-controls on most bleeding dates. Lowest serum albumin concentrations occurred in the surviving pig of group 9 (16 mg/kg lead acetate i.p.).

A significant ($P < .05$) increase of the mean serum albumin concentration for the entire trial was found in group 2 (16 mg/kg lead acetate orally), and weekly mean serum albumin concentrations of this group were higher than those of the oral-control group on most sampling days.

No significant group differences in mean serum $alpha_1$ globulin concentrations for the whole experiment were noted.

Significant ($P < .05$) and highly significant ($P < .01$) elevations of mean serum $alpha_2$ globulin concentrations for the entire experiment were found in groups 6 (2 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.), respectively; but a significant ($P < .05$) reduction of this same value was detected in group 3 (32 mg/kg lead acetate orally). Weekly

mean serum $alpha_2$ globulin concentrations fluctuated in all groups during the experiment but highest values were found in group 9 and lowest values were noted in group 3.

Mean serum $beta_1$ globulin concentrations for the whole trial were significantly ($P < .05$) elevated in groups 3 (32 mg/kg lead acetate orally) and 8 (8 mg/kg lead acetate i.p.). Highest weekly mean serum $beta_1$ globulin concentrations in orally-exposed groups occurred in group 3 while those in i.p.-exposed groups occurred in group 8.

Significantly ($P < .05$) decreased mean serum $beta_2$ globulin concentrations for the whole experiment were found in groups 3 (32 mg/kg lead acetate orally) and 4 (64 mg/kg lead acetate orally).

Significant ($P < .05$) increases in mean serum gamma globulin concentrations and albumin:globulin ratios for the entire experiment occurred in group 4 (64 mg/kg lead acetate orally), and highly significant ($P < .01$) increases of these same values occurred in groups 7 (4 mg/kg lead acetate i.p.), 8 (8 mg/kg lead acetate i.p.), and 9 (16 mg/kg lead acetate i.p.). Weekly mean serum gamma globulin concentrations of group 4 were higher than those of other orally lead or sodium acetate-exposed groups on most bleeding dates. After the fifth week of lead acetate exposure, weekly serum gamma globulin concentrations of groups 7, 8, and 9 were higher than those of i.p.-controls on most bleeding dates.

Mean total serum protein, albumin, $alpha_1$ globulin, $alpha_2$ globulin, $beta_1$ globulin, $beta_2$ globulin, and gamma globulin concentrations and albumin:globulin ratios are presented in Tables 9 and 10. Results of statistical analyses are also summarized in these tables. Weekly group means are given in the Appendix (Tables 68-83).

Tissue lead concentrations

Tissue lead concentrations in orally lead acetate-exposed pigs were consistently greatest in the rib. Bone marrow, kidney, liver, spleen, and lung all had high lead concentrations in relation to other organs in various groups. Lymph node, skeletal muscle, and brain had consistently lower lead concentrations.

In i.p. lead acetate-exposed pigs, greatest tissue lead concentrations were consistently found in the rib with lymph node, lung, liver, spleen, and bone marrow also having very high concentrations. Kidney lead concentrations were markedly elevated but were lower than concentrations in the previously mentioned organs. Lead concentrations in skeletal muscle and brain were very low.

Mean tissue lead concentrations tended to fall in lead acetate-exposed pigs during the 60 day period after cessation of lead acetate exposure. This fall was not noted in tissues which had accumulated only small amounts of lead such as

Table 9. Experiment 1--Orally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 1 | 2 | 3 | 4 | 5 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Serum concentrations[c] | | | | | | | |
| Total protein | 6.3 | 6.7 | 5.9* | 6.2 | 6.4 | 0.4 | 0.6 |
| Albumin | 2.65 | 2.91* | 2.35** | 2.30** | 2.68 | 0.18 | 0.26 |
| Globulins | | | | | | | |
| $Alpha_1$ | 0.26 | 0.23 | 0.21 | 0.24 | 0.25 | 0.05 | 0.07 |
| $Alpha_2$ | 0.97 | 1.08 | 0.86* | 1.02 | 1.04 | 0.13 | 0.18 |
| $Beta_1$ | 0.35 | 0.32 | 0.40* | 0.30 | 0.31 | 0.07 | 0.10 |
| $Beta_2$ | 0.93 | 0.96 | 0.87* | 0.88* | 1.02 | 0.13 | 0.18 |
| Gamma | 1.16 | 1.15 | 1.24 | 1.45* | 1.11 | 0.33 | 0.46 |
| Albumin:globulin ratio | 0.73 | 0.78 | 0.66 | 0.60* | 0.72 | 0.09 | 0.12 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 1, 2, 3, and 4 received 8, 16, 32, and 64 mg/kg lead acetate, respectively.

[c]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group 5.

**Highly significant difference ($P < .01$) as compared to group 5.

Table 10. Experiment 1--Intraperitoneally lead or sodium acetate-exposed groups. Means[a] for the entire experiment.

| Measurement | Group[b] 6 | 7 | 8 | 9 | 10 (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|---|---|
| Serum concentrations[c] | | | | | | | |
| Total protein | 6.3 | 6.5 | 6.8 | 6.2 | 6.6 | 0.5 | 0.8 |
| Albumin | 2.42** | 2.22** | 2.15** | 1.84** | 2.65 | 0.20 | 0.30 |
| Globulins | | | | | | | |
| $Alpha_1$ | 0.22 | 0.25 | 0.27 | 0.22 | 0.24 | 0.05 | 0.07 |
| $Alpha_2$ | 1.10* | 1.06 | 1.04 | 1.27** | 1.02 | 0.07 | 0.11 |
| $Beta_1$ | 0.38 | 0.39 | 0.51* | 0.44 | 0.35 | 0.12 | 0.17 |
| $Beta_2$ | 0.89 | 0.89 | 1.00 | 0.87 | 0.98 | 0.14 | 0.20 |
| Gamma | 1.27 | 1.73** | 1.75** | 1.57* | 1.28 | 0.23 | 0.34 |
| Albumin:globulin ratio | 0.64 | 0.52** | 0.47** | 0.42** | 0.68 | 0.07 | 0.11 |

[a]Bleeding prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups 6, 7, 8, and 9 received 2, 4, 8, and 16 mg/kg lead acetate, respectively.

[c]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group 10.

**Highly significant difference ($P < .01$) as compared to group 10.

brain and skeletal muscle. Lead concentrations of lung actually rose during the 60 day post-lead-exposure period in pigs which had been exposed to lead acetate orally. It is also notable that highest lead concentrations in rib and bone marrow occurred in both orally and i.p. lead acetate-exposed pigs euthanatized 30 days after lead exposure had ceased.

Mean lead concentrations in tissues from orally and i.p. lead and sodium acetate-exposed groups are given in Tables 11 and 12. Mean tissue lead concentrations of orally lead acetate-exposed, i.p. lead acetate-exposed, and sodium acetate-control pigs euthanatized at 1, 30, or 60 days after cessation of lead or sodium acetate exposure are given in Table 13.

Macroscopic lesions

Few significant macroscopic lesions were observed in pigs which received lead or sodium acetate orally. Adhesions were present in the pleural and pericardial cavities of 2 pigs from group 4 (64 mg/kg lead acetate orally).

Adhesions, which were centered at injection sites, were observed in abdominal cavities of all but 1 i.p. lead acetate-exposed pig. Only small portions of intestine were involved in groups 6 (2 mg/kg lead acetate i.p.) and 7 (4 mg/kg lead acetate i.p.). In groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.) larger

Table 11. Experiment 1--Orally lead or sodium acetate-exposed groups. Mean[a] tissue lead concentrations (ppm).

| Tissue | Group[b] 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| Kidney | 4.85 | 5.92 | 4.74 | 9.00 | 1.34 |
| Liver | 5.42 | 3.18 | 3.84 | 4.77 | 2.25 |
| Spleen | 5.52 | 4.83 | 3.21 | 1.40 | 2.00 |
| Lymph node | 2.50 | 3.25 | 1.50 | 1.80 | 0.63 |
| Lung | 4.77 | 15.35 | 3.48 | 113.75 | 0.67 |
| Brain | 1.77 | 1.52 | 1.54 | 0.97 | 0.79 |
| Skeletal muscle | 1.05 | 1.60 | 0.95 | 0.87 | 0.59 |
| Rib | 44.09 | 63.09 | 336.74 | 356.33 | 15.03 |
| Bone marrow | 5.25 | 8.18 | 4.67 | 2.55 | 0.58 |

[a]Means of 3 pigs euthanatized at 1, 30, and 60 days after cessation of lead or sodium acetate exposure.

[b]Groups 1, 2, 3, and 4 received 8, 16, 32, and 64 mg/kg lead acetate, respectively.

Table 12. Experiment 1--Intraperitoneally lead or sodium acetate-exposed pigs. Mean tissue lead concentrations (ppm).

| Tissue | Group[a] | | | | |
|---|---|---|---|---|---|
| | 6[b] | 7[b] | 8 | 9 | 10[b] (control) |
| Kidney | 6.67 | 8.67 | 7.00[c] | 9.14[c] | 3.00 |
| Liver | 6.00 | 10.17 | 40.75[c] | 30.33[c] | 2.25 |
| Spleen | 3.60 | 4.43 | 18.80[d] | 89.83[c] | 3.75 |
| Lymph node | 25.30 | 13.75 | 385.00[d] | --[e] | 3.38 |
| Lung | 9.30 | 19.75 | 31.00[d] | 15.25[d] | 2.00 |
| Brain | 2.18 | 2.04 | 1.20[c] | 3.09[c] | 1.50 |
| Skeletal muscle | 1.27 | 1.04 | 0.90[c] | 0.75[c] | 2.07 |
| Rib | 191.85 | 497.64 | 641.90[d] | 941.80[d] | 17.41 |
| Bone marrow | 8.77 | 22.63 | 18.00[d] | 27.50[d] | 2.00 |

[a]Groups 6, 7, 8, and 9 received 2, 4, 8, and 16 mg/kg lead acetate, respectively.

[b]Mean of 3 pigs euthanatized at 1, 30, and 60 days after cessation of lead or sodium acetate exposure.

[c]Mean concentrations from 2 pigs dying during the experiment and 1 pig which survived to the end of the experiment.

[d]Mean from the pig in this group which survived to the end of the experiment.

[e]Three different lymph nodes examined:
Mesenteric = 31.80 ppm
Cervical = 2.00 ppm
Prefemoral = 1050.00 ppm

Table 13. Experiment 1--Mean lead concentrations of selected tissues on 1, 30, and 60 days after cessation of lead or sodium acetate exposure.

| Tissue | Orally lead-exposed[a] | | | Intraperitoneally lead-exposed[b] | | | Oral- and intraperitoneal-control[c] | | |
|---|---|---|---|---|---|---|---|---|---|
| | Days | | | | | | | | |
| | 1 | 30 | 60 | 1 | 30 | 60 | 1 | 30 | 60 |
| Kidney | 10.12 | 3.58 | 3.69 | 10.25 | 8.12 | 4.62 | 3.00 | 0.75 | 1.62 |
| Liver | 7.02 | 3.63 | 2.25 | 9.25 | 10.00 | 5.00 | 1.38 | 2.25 | 1.75 |
| Spleen | 5.48 | 2.40 | 3.60 | 6.40 | 3.52 | 2.12 | 2.50 | 3.00 | 2.75 |
| Lymph node | 2.73 | 1.00 | 4.75 | 47.00 | 9.90 | 1.68 | 1.44 | 0.50 | 2.62 |
| Lung | 13.32 | 16.25 | 49.06 | 27.00 | 7.82 | 9.50 | 0.69 | 0.88 | 1.88 |
| Brain | 1.30 | 1.25 | 1.80 | 1.69 | 2.20 | 2.44 | 0.69 | 0.50 | 1.75 |
| Skeletal muscle | 0.93 | 1.40 | 1.01 | 0.50 | 1.15 | 1.82 | 1.13 | 0.50 | 1.56 |
| Rib | 187.68 | 255.06 | 97.69 | 229.60 | 388.17 | 282.45 | 13.28 | 21.92 | 16.40 |
| Bone marrow | 5.80 | 6.09 | 4.49 | 4.20 | 24.05 | 9.62 | 1.38 | 1.00 | 1.00 |

[a]Mean of 4 orally lead-exposed pigs euthanatized on each of these days.

[b]Mean of 2 pigs, 1 each from groups 6 and 7, euthanatized on each of these days.

[c]Mean of 1 oral control and 1 i.p. control pig euthanatized on each of these days.

sections of intestine and other abdominal organs were adhered to each other and to the parietal peritoneum.

During the experiment, 4 pigs exposed to lead acetate i.p. died. Partial to complete intestinal obstruction, gas- and fluid-filled intestines cranial to these obstructions, and 1 intussuception were observed in these pigs.

No macroscopic lesions were found in oral or i.p. sodium acetate-control pigs.

Microscopic lesions

Slight to moderate extramedullary hematopoiesis was evident in livers and kidneys of many orally lead or sodium acetate-exposed pigs, and interstitial foci of lymphocytes were present in kidneys of a few of these pigs.

Lymphoid hyperplasia adjacent to bronchioles and slight thickening of alveolar walls with fibrous connective tissue were observed in lungs of orally lead acetate-exposed pigs, but not in those of oral sodium acetate-controls. A chronic diffuse pleuritis characterized by fibrin, macrophages, and villous proliferation of the visceral pleura was evident in 2 pigs from group 4 (64 mg/kg lead acetate orally).

A chronic diffuse epicarditis characterized by proliferation of fibrous connective tissue was detected in 2 pigs from group 4 (64 mg/kg lead acetate orally).

No attempt was made to isolate bacterial or mycoplasmal organisms.

Large, eosinophilic, acid-fast, intranuclear inclusion bodies were visualized in hepatocytes and epithelial cells of proximal renal tubules in 1 pig from group 4 (64 mg/kg lead acetate orally) but not in tissues of oral sodium acetate-control pigs.

Foci of extramedullary hematopoiesis and/or mononuclear inflammatory cells were found in livers and kidneys of both i.p. lead and sodium acetate-exposed pigs.

Lymphoid hyperplasia adjacent to bronchioles was evident in lungs of both i.p. lead and sodium acetate-exposed pigs.

Numerous large, eosinophilic, acid-fast, intranuclear inclusion bodies were present in hepatocytes of surviving pigs from groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.). A few small, eosinophilic, acid-fast, intranuclear inclusion bodies were observed in epithelial cells of proximal renal tubules of the surviving pig from group 8. Inclusions were not observed in tissues of i.p. sodium acetate-control pigs.

Chronic granulomatous lymphadenitis characterized by R-E hyperplasia and a few foreign-body giant cells was evident in lymph nodes of 2 pigs from group 9 (16 mg/kg lead acetate i.p.). The granulomatous reaction was most intense adjacent

to the hilus of the nodes. An amorphous, yellow-brown birefringent material was present both within R-E cells and extracellularly. This material was most dense near the hilus and was mineralized in this area.

Chronic focal granulomatous peritonitis characterized by well encapsulated masses containing lymphocytes, macrophages, and foreign-body giant cells and/or subacute focal to diffuse, fibrinous peritonitis characterized by accumulations of fibrin, macrophages, and neutrophils were observed in i.p. lead acetate-exposed pigs. An amorphous, yellow-brown, birefringent material was present both within phagocytic cells and extracellularly in the granulomatous masses. This material was partially mineralized. In a few pigs, organization of fibrin on serosal surfaces and/or mesothelial proliferation were visualized.

No microscopic lesions were observed in spleen, cerebrum, cerebellum, spinal cord, or ischiatic nerve of pigs exposed to lead or sodium acetate either orally or i.p.

## Experiment 2

### Clinical signs and mortalities

No clinical abnormalities were observed in lead or sodium acetate-exposed pigs during the pre-challenge period.

Following i.p. challenge with Salmonella, clinical signs were evident in 12 of the 18 pigs previously exposed to lead.

Onset of illness was consistently on the second day after challenge. Signs varied from slight inappetence and depression to complete anorexia, weakness, gauntness, shivering, cyanosis of extremities, increased respiratory rates, and yellow, watery diarrhea. Duration of clinical signs was 1-9 days. Four of the 18 pigs were judged to be severely ill. One pig developed cyanosis of extremities, and 4 pigs had periods of diarrhea. No mortalities occurred in lead acetate-exposed replicates.

Clinical signs were observed in 11 of 18 sodium acetate-exposed pigs following challenge. Signs were similar to those observed in lead acetate-exposed pigs. Time of onset was again 2 days, and duration of illness varied from 1-9 days. Seven of the 18 pigs were severely ill. Five pigs developed cyanosis of extremities, and 6 had diarrhea. One pig from the sodium acetate-exposed group died. This pig had severe cyanosis but did not develop diarrhea.

Body temperatures

Mean body temperatures of lead or sodium acetate-exposed groups were not significantly different during either the pre- or post-challenge period. Prior to Salmonella challenge, mean body temperatures of all replicates remained within normal limits ($38.6^{\circ}$-$40.0^{\circ}$C) but were above normal by the third day after challenge and remained elevated for 4-6 days.

Mean body temperatures prior to and following challenge and results of statistical analysis are presented in Table 14. Daily group means are given in the Appendix (Tables 84 and 85).

Table 14. Experiment 2--Group means[a] from 3 replicates.

| Measurement | Group A (lead-exposed) | Group B (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|
| Body temperature (°C) | | | | |
| Pre-challenge[b] | 39.6 | 39.5 | 0.4 | 0.6 |
| Post-challenge[c] | 40.2 | 40.3 | 0.4 | 0.6 |
| Blood lead concentration (ug/dl) | 33** | 4 | 2 | 4 |
| Blood ALAD activity | 42.7** | 200.3 | 41.7 | 70.7 |
| Hematocrit (%) | | | | |
| Pre-challenge[b] | 33 | 32 | 2 | 3 |
| Post-challenge[c] | 31 | 30 | 2 | 3 |

[a]Bleedings prior to lead or sodium acetate exposure not used in calculating means.

[b]Period of lead or sodium acetate exposure prior to *Salmonella* challenge.

[c]Following *Salmonella* challenge.

**Highly significant difference ($P < .01$) as compared to group B.

Blood lead concentrations

A highly significant ($P < .01$) increase in mean blood lead concentration for the entire trial occurred in group A (lead acetate-exposed) as compared to group B (sodium acetate-exposed).

Prior to initiation of lead or sodium acetate exposure, the mean blood lead concentration was 6 ug/dl. Mean blood lead concentrations of the 3 lead acetate-exposed replicates at the time of challenge were 42, 42, and 48 ug/dl. In the 3 sodium acetate-exposed replicates, mean blood lead concentrations at the time challenge were 1, 5, and 2 ug/dl.

Mean blood lead concentrations and results of statistical analysis are given in Table 14. Daily group means are summarized in the Appendix (Table 86).

Blood ALAD activities

A highly significant ($P < .01$) decrease in mean blood ALAD activity for the whole experiment was detected in group A (lead acetate-exposed) as compared to group B (sodium acetate-exposed).

Mean blood ALAD activity prior to the beginning of lead or sodium acetate exposure was 219.3 units. Blood ALAD activities decreased rapidly during the first 7 days in lead-exposed pigs. Activities of ALAD remained high in sodium acetate-exposed pigs during the first 2 weeks of such exposure but fell slightly during the third week.

Mean blood ALAD activities and results of statistical analysis are summarized in Table 14. Daily group means are presented in the Appendix (Table 87).

Hematologic observations

No significant group differences in mean hematocrit percentages were detected during either the pre- or post-challenge period. Hematocrit percentages prior to challenge were normal (32-50%) except in replicate 1 of group B (sodium acetate-exposed) where this value was slightly low (31%). Hematocrit percentages had fallen below normal in all replicates by the end of the post-challenge period.

Mean hematocrit percentages, prior to and following challenge, and results of statistical analysis are given in Table 14. Daily group means are presented in the Appendix (Table 88).

The erythrocyte morphology of lead and sodium acetate-exposed pigs was not significantly different. Variable anisocytosis and polychromasia of erythrocytes were observed during the trial. Occasional nucleated erythrocytes were also present in blood smears. Basophilic stippling was not observed.

No significant group differences in mean total leukocyte counts for either the pre- or post-challenge period were noted. Prior to Salmonella challenge, mean total leukocyte

counts were normal (11,000-22,000/cubic mm) except in replicate 3 of group B (sodium acetate-exposed) where slight elevations occurred. By 2 days post-challenge, total leukocyte counts were above normal in all replicates except replicate 2 of group A (lead acetate-exposed). Mean total leukocyte counts had fallen by the fourth day after challenge and were normal in all replicates by the ninth day of the post-challenge period.

The mean total segmented neutrophil count of group A (lead acetate-exposed) was significantly ($P < .05$) lower than that of group B (sodium acetate-exposed) during the pre-challenge period, but no significant group differences were detected following Salmonella challenge. Total segmented neutrophil counts were above normal (3,200-10,000/cubic mm) in 1 lead and 2 sodium acetate-exposed replicates and below normal in 1 lead acetate-exposed replicate on the second day after challenge, but were within normal limits by 4 days after challenge.

Significant group differences in total band neutrophil counts were not found during the pre-challenge period, but following Salmonella challenge, the mean total band neutrophil count in group A (lead acetate-exposed) was significantly ($P < .05$) lower than that of group B (sodium acetate-exposed). Prior to challenge, band neutrophil counts were above normal (0-800/cubic mm) in 1 lead and 2 sodium

acetate-exposed replicates. Band neutrophil counts had risen sharply by 2 days after challenge and, although they remained above normal, fell gradually in most replicates during the remainder of the post-challenge period.

Mean total lymphocyte, eosinophil, and monocyte counts of group A (lead acetate-exposed) were not significantly different from those of group B (sodium acetate-exposed) during either the pre- or post-challenge period.

On certain sampling dates during the pre-challenge period, total lymphocyte counts were above normal (4,500-13,000/cubic mm) in 1 lead and 1 sodium acetate-exposed replicate. After Salmonella challenge, total lymphocytes counts of all replicates decreased, but counts fell below normal only in 1 sodium acetate-exposed replicate.

Eosinophil counts were lowest on the second day after Salmonella challenge in most replicates, but counts fell below normal (50-2,000/cubic mm) in only 2 lead and 1 sodium acetate-exposed replicate. Eosinophil counts had risen by the fourth day after challenge.

Mean plasma protein:fibrinogen ratios of groups A (lead acetate-exposed) and B (sodium acetate-exposed) were not significantly different during either the pre- or post-challenge period. Ratios had decreased by the second day after Salmonella challenge and stayed low in most replicates throughout the post-challenge period.

Mean total leukocyte, segmented neutrophil, band neutrophil, lymphocyte, eosinophil, and monocyte counts and mean plasma protein:fibrinogen ratios, prior to and following challenge, are given in Tables 15 and 16. Results of statistical analyses are also summarized in these tables. Daily group means are presented in the Appendix (Tables 89-95).

Agarose film electrophoresis

Significant group differences in mean total serum protein concentrations were not found during either the pre- or post-challenge periods. Mean total serum protein concentrations of all replicates rose after challenge and remained high during the post-challenge period.

Mean serum albumin concentrations of groups A (lead acetate-exposed) and B (sodium acetate-exposed) were not significantly different during either the pre- or post-challenge period. Concentrations decreased in all replicates following Salmonella challenge.

There were no significant group differences in serum $alpha_1$ globulin concentrations during either the pre- or post-challenge period. In most replicates, concentrations decreased gradually after Salmonella challenge.

Mean serum $alpha_2$ globulin concentrations of groups A (lead acetate-exposed) and B (sodium acetate-exposed) were not significantly different during the period before

Table 15. Experiment 2--Group means[a] from 3 replicates. Period of lead or sodium acetate exposure prior to *Salmonella* challenge.

| Measurement | Group A (lead-exposed) | Group B (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|
| *Total counts*[b] | | | | |
| Leukocyte | 17,626 | 20,384 | 2,884 | 4,645 |
| Segmented neutrophil | 4,859* | 7,428 | 2,300 | 3,625 |
| Band neutrophil | 446 | 513 | 1,112 | 1,758 |
| Lymphocyte | 11,842 | 12,024 | 1,090 | 1,648 |
| Eosinophil | 382 | 331 | 182 | 297 |
| Monocyte | 138 | 82 | 131 | 204 |
| Plasma protein: fibrinogen ratio | 25 | 21 | 7 | 11 |

[a]Bleedings prior to lead or sodium acetate exposure not used in calculating means.

[b]Cells/cubic mm of blood.

*Significant difference ($P < .05$) as compared to group B.

*Salmonella* challenge, but during the post-challenge period, the mean concentration of group A was significantly ($P < .05$) lower than that of group B. While mean $alpha_2$ globulin concentrations of both groups rose following challenge, the rise was greater in group B (sodium acetate-exposed).

Table 16. Experiment 2--Group means from 3 replicates. Period following *Salmonella* challenge.

| Measurement | Group A (lead-exposed) | Group B (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|
| *Total counts*[a] | | | | |
| Leukocyte | 21,733 | 22,580 | 2,884 | 4,645 |
| Segmented neutrophil | 6,619 | 6,187 | 2,300 | 3,625 |
| Band neutrophil | 6,559* | 8,159 | 1,112 | 1,758 |
| Lymphocyte | 8,205 | 7,858 | 1,090 | 1,648 |
| Eosinophil | 293 | 275 | 182 | 297 |
| Monocyte | 89 | 96 | 131 | 204 |
| Plasma protein: fibrinogen ratio | 17 | 17 | 7 | 11 |

[a]Cells/cubic mm of blood.

*Significant difference ($P < .05$) as compared to group B.

During the period prior to *Salmonella* challenge, the mean serum $beta_1$ globulin concentration of group A (lead acetate-exposed) was significantly ($P < .05$) higher than that of group B (sodium acetate-exposed). Following challenge, no significant group difference in mean $beta_1$ globulin concentration was noted. Serum $beta_1$ globulin concentrations rose in all replicates following challenge.

No significant group differences in mean $beta_2$ globulin concentrations were found during either the pre- or post-challenge period.

Significant group differences in mean serum gamma globulin concentrations were not detected during either the pre- or post-challenge period. Gamma globulin concentrations of all replicates rose gradually throughout the trial.

The mean albumin:globulin (A/G) ratio of group A (lead acetate-exposed) was not significantly different from that of group B (sodium acetate-exposed) during the pre-challenge period. Following Salmonella challenge, the mean A/G ratio of group A was significantly ($P < .05$) higher than that of group B. Albumin:globulin ratios of all replicates decreased following challenge.

Mean total serum protein, albumin, $alpha_1$ globulin, $alpha_2$ globulin, $beta_1$ globulin, $beta_2$ globulin, and gamma globulin concentrations and mean albumin:globulin ratios, prior to and following challenge, are presented in Tables 17 and 18. Results of statistical analyses are also summarized in these tables. Daily group means are given in the Appendix (Tables 96-103).

Serum agglutinin titrations

The mean serum S. choleraesuis var. Kunzendorf somatic agglutinin titer of group A (lead acetate-exposed) was significantly ($P < .05$) higher than that of group B (sodium

Table 17. Experiment 2--Group means[a] from 3 replicates. Period of lead or sodium acetate exposure prior to Salmonella challenge.

| Measurement | Group A (lead-exposed) | Group B (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|
| Serum concentrations[b] | | | | |
| Total protein | 6.2 | 6.0 | 0.3 | 0.5 |
| Albumin | 2.25 | 2.15 | 0.12 | 0.19 |
| Globulins | | | | |
| $Alpha_1$ | 0.34 | 0.33 | 0.06 | 0.09 |
| $Alpha_2$ | 1.24 | 1.19 | 0.07 | 0.11 |
| $Beta_1$ | 0.48* | 0.43 | 0.05 | 0.07 |
| $Beta_2$ | 0.91 | 0.83 | 0.09 | 0.13 |
| Gamma | 1.01 | 1.04 | 0.18 | 0.29 |
| Albumin:globulin ratio | 0.56 | 0.57 | 0.02 | 0.03 |

[a]Bleedings prior to lead or sodium acetate exposure not used in calculating means.

[b]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group B.

Table 18. Experiment 2--Group means from 3 replicates. Period following Salmonella challenge.

| Measurement | Group A (lead-exposed) | Group B (control) | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|
| Serum concentrations[a] | | | | |
| Total protein | 6.8 | 6.8 | 0.3 | 0.5 |
| Albumin | 2.03 | 2.03 | 0.12 | 0.19 |
| Globulins | | | | |
| $Alpha_1$ | 0.33 | 0.34 | 0.06 | 0.09 |
| $Alpha_2$ | 1.61* | 1.69 | 0.07 | 0.11 |
| $Beta_1$ | 0.63 | 0.67 | 0.05 | 0.07 |
| $Beta_2$ | 0.86 | 0.83 | 0.09 | 0.13 |
| Gamma | 1.29 | 1.27 | 0.18 | 0.29 |
| Albumin:globulin ratio | 0.43* | 0.41 | 0.02 | 0.03 |

[a]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group B.

acetate-exposed) on the ninth day after Salmonella challenge. No other significant group differences were noted on either the day prior to challenge or on days 2, 4, or 7 following challenge.

There were no significant group differences in mean serum S. choleraesuis var. Kunzendorf flagellar agglutinin titers on the day prior to Salmonella challenge or on days 2, 4, 7, or 9 following challenge.

Daily mean somatic and flagellar agglutinin titers on the day prior to and on several days following challenge are summarized in Tables 19 and 20.

Serum complement titrations

Mean serum complement titers for the pre-challenge period were 95.2 and 89.9 in groups A (lead acetate-exposed) and B (sodium acetate-exposed), respectively. Following Salmonella challenge, mean serum complement titers were 112.4 and 101.1 in groups A and B, respectively. Group differences were not found to be significant.

Daily mean serum complement titers for the pre- and post-challenge periods are summarized in Tables 21 and 22.

Salmonella isolation from tissues

Salmonella organisms were most frequently isolated from the mesenteric lymph node and ileum. While Salmonella organisms were isolated from 12 of 108 tissues from group A (lead acetate-exposed), they were isolated from 27 of 108 from group B (sodium acetate-exposed). Organisms were isolated from 7 of 18 lead acetate-exposed pigs and from 10

Table 19. Experiment 2--Geometric means of S. choleraesuis var. Kunzendorf somatic agglutinin titers on a daily basis.

| | Group A (lead-exposed) | | | | Group B (control) | | | |
|---|---|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Mean | Rep 1 | Rep 2 | Rep 3 | Mean |
| 0[a] | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 4 | 7 | 0 | 4 | 4 | 0 | 1 | 3 | 1 |
| 7 | 9 | 13 | 14 | 12 | 3 | 15 | 13 | 10 |
| 9 | 28 | 20 | 28 | 25* | 11 | 15 | 18 | 15 |

[a]Day of Salmonella challenge.

*Significantly different ($P < .05$) as compared to the mean of group B on this day.

Table 20. Experiment 2--Geometric means of S. choleraesuis var. Kunzendorf agglutin titers on a daily basis.

| | Group A (lead-exposed) | | | | Group B (control) | | | |
|---|---|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Mean | Rep 1 | Rep 2 | Rep 3 | Mean |
| 0[a] | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 2 | 0 | 0 | 0 | 0 | 0 | 0 | 0 | 0 |
| 4 | 20 | 35 | 80 | 45 | 25 | 40 | 160 | 75 |
| 7 | 870 | 1,955 | 2,280 | 1,700 | 1,610 | 5,110 | 6,450 | 4,390 |
| 9 | 2,030 | 6,450 | 4,560 | 4,350 | 2,880 | 13,500 | 14,500 | 10,295 |

[a]Day of Salmonella challenge.

Table 21. Experiment 2--Mean serum complement ($CH_{50}$) titers during the lead or sodium acetate-exposure period prior to Salmonella challenge.

| Day | Group A (lead-exposed) | Group B (control) |
|---|---|---|
| 0[a] | 76.9 | 75.9 |
| 2 | 103.2 | 91.5 |
| 4 | 95.6 | 93.9 |
| 8 | 93.9 | 96.3 |
| 15[b] | 89.4 | 86.6 |
| 21[c] | 111.9 | 95.1 |

[a]Prior to lead or sodium acetate exposure.

[b]Means from replicates 2 and 3.

[c]Means from replicate 3 only.

Table 22. Experiment 2--Mean serum complement ($CH_{50}$) titers following Salmonella challenge.

| Day | Group A (lead-exposed) | Group B (control) |
|---|---|---|
| 2 | 116.4 | 104.5 |
| 4 | 126.3 | 113.1 |
| 7 | 100.6 | 95.1 |
| 9 | 105.9 | 91.8 |

of 18 sodium acetate-exposed pigs. All isolates were serotyped as S. choleraesuis var. Kunzendorf.

Results of attempted Salmonella isolation from various tissues are given in Table 23.

Tissue lead concentrations

The mean lead concentrations of livers and kidneys from group A (lead acetate-exposed) were 100% and 75% higher, respectively, than mean concentrations of kidneys and livers from group B (sodium acetate-exposed). The mean lead concentration of lymph nodes from group B was 20% higher than that of lymph nodes from group A. Mean brain lead concentrations of the 2 groups were very similar.

Mean lead concentrations of various tissues are summarized in Table 24.

Macroscopic lesions

Focal areas of the mucosal surface of the cecum, colon, and/or small intestine were unusually red in about 50% of the pigs (9 of 18 lead acetate-exposed; 8 of 18 sodium acetate-exposed). Yellow raised nodules (button ulcers) were evident on the mucosal surface of the cecum of 1 sodium acetate-exposed pig but were not observed in pigs exposed to lead acetate.

Spleens of some pigs (4 of 18 lead acetate-exposed; and 4 of 18 sodium acetate-exposed) were enlarged 2-4 times and

Table 23. Experiment 2--Positive Salmonella isolates from selected tissues/number of pigs in replicate or group.

| Group | Tissue | | | | | |
|---|---|---|---|---|---|---|
| | Kidney | Liver | Spleen | Mesenteric lymph node | Mandibular lymph node | Ileum |
| A (lead-exposed) | | | | | | |
| Rep 1 | 0/6 | 0/6 | 1/6 | 2/6 | 1/6 | 0/6 |
| Rep 2 | 0/6 | 0/6 | 0/6 | 0/6 | 0/6 | 0/6 |
| Rep 3 | 0/6 | 0/6 | 0/6 | 2/6 | 2/6 | 4/6 |
| Total | 0/18 | 0/18 | 1/18 | 4/18 | 3/18 | 4/18 |
| B (control) | | | | | | |
| Rep 1 | 1/6 | 2/6 | 2/6 | 4/6 | 3/6 | 3/6 |
| Rep 2 | 1/6 | 1/6 | 1/6 | 2/6 | 1/6 | 3/6 |
| Rep 3 | 0/6 | 0/6 | 0/6 | 1/6 | 0/6 | 2/6 |
| Total | 2/18 | 3/18 | 3/18 | 7/18 | 4/18 | 8/18 |

Table 24. Experiment 2--Mean tissue lead concentrations (ppm) of selected tissues.

| Group | Tissue | | | |
|---|---|---|---|---|
| | Kidney | Liver | Lymph node | Brain |
| A (lead-exposed) | | | | |
| Rep 1 | 3.25 | 3.83 | 2.17 | 1.75 |
| Rep 2 | 3.50 | 3.75 | 2.00 | 2.00 |
| Rep 3 | 3.75 | 4.50 | 2.00 | 1.58 |
| Total | 3.50 | 4.03 | 2.06 | 1.78 |
| B (control) | | | | |
| Rep 1 | 1.83 | 1.49 | 2.92 | 1.62 |
| Rep 2 | 2.67 | 2.17 | 2.08 | 2.04 |
| Rep 3 | 2.00 | 2.50 | 2.28 | 1.38 |
| Total | 2.17 | 2.05 | 2.43 | 1.68 |

dark red. Focal deposits of white, fibrinous material were present on serosal surfaces of abdominal organs in 50% of the pigs (10 of 18 lead acetate-exposed; 8 of 18 sodium acetate-exposed).

Microscopic lesions

Acute multifocal hepatitis characterized by coagulative necrosis with no definite lobular distribution surrounded by accumulations of lymphoreticular cells and neutrophils, moderate hyperplasia of Kupffer cells, and increased neutrophil numbers in sinusoids was evident in 17 of 36 pigs (8 of 18 lead acetate-exposed; 9 of 18 sodium acetate-exposed).

Splenitis characterized by multifocal to diffuse neutrophil accumulations and lymphoid depletion was detected in nearly all spleens from both lead and sodium acetate-exposed pigs. A small number of spleens were congested.

Acute multifocal to diffuse interstitial pneumonia characterized by accumulations of neutrophils and macrophages in alveolar walls was found in nearly all lead or sodium acetate-exposed pigs. An acute fibrinous pneumonia characterized by the presence of fibrin and macrophages in alveoli and bronchioles was evident in the 1 pig which died during the course of the experiment.

Lymphoid depletion was observed in most lymph nodes. Lymphadenitis characterized by edema, microabscesses, and focal coagulative necrosis was found in mesenteric lymph nodes of a small number of pigs.

Increased numbers of mononuclear inflammatory cells were present in the lamina propria and submucosa, and increased mucin-producing cells were present in the mucosa of the ileum, colon, and cecum of most pigs. Coagulative necrosis involving the mucosa and submucosa of the cecum was evident in 1 sodium acetate-exposed pig. Macrophages, eosinophils, and neutrophils were present in tissues adjacent to the necrotic area.

Chronic multifocal fibrinous peritonitis characterized by fibrin, macrophages, and neutrophils plus variable amounts of loose connective tissue and fibroblasts was detected in most lead and sodium acetate-exposed pigs.

No microscopic lesions were observed in kidneys.

## Experiment 3

### Clinical signs and mortalities

No clinical signs of overt lead toxicosis were noted in either of the 2 lead chloride-exposed groups; however, appetites of pigs in group B appeared to be somewhat decreased near the end of the 2 week lead chloride-exposure period. No other clinical abnormalities were detected in the 3 groups prior to challenge.

During the first 24 hours after challenge, all pigs appeared to be normal. By 48 hours post-challenge, about 50% of the pigs were visibly depressed, a smaller number were

unusually gaunt, and a few were suffering from diarrhea and/or vomition. At 72 hours, their conditions varied from normal to extremely depressed and gaunt. All were anorectic, about 40% were suffering from diarrhea, and a few were visibly dehydrated. During the next 11 days, the conditions of all pigs deteriorated. Most continued to have diarrhea until death or until the termination of the experiment. Appetites, while quite variable, seemed to return to normal in animals surviving after 8 days post-challenge.

The severity of clinical signs was not judged to be visibly different between groups. No statistically significant group differences were noted in the 4 numerically rated categories of clinical signs. Mean scores for each of these categories are summarized in Table 25.

Table 25. Experiment 3--Group means for 4 categories of numerically-rated clinical signs following *Salmonella* challenge.

| Group[a] | Category | | | |
|---|---|---|---|---|
| | General condition | Hydration | Appetite | Fecal consistency |
| A(control) | 1.8 | 1.4 | 1.6 | 2.1 |
| B | 1.6 | 1.4 | 1.7 | 1.8 |
| C | 1.7 | 1.3 | 1.6 | 2.0 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

The first death occurred in group C (1 week lead chloride) on the third day post-challenge. All pigs in group B (2 weeks lead chloride) were dead by 12 days after challenge. Seven pigs from group C and 4 pigs from group A (sodium chloride-control) had died by the time the trial was terminated at 14 days post-challenge. Daily and cumulative mortality rates are summarized in Figure 1. While 15 of 16 (93.8%) lead chloride-exposed pigs died, 4 of 8 (50.0%) sodium chloride-control pigs died. The mortality rate in lead chloride-exposed pigs was significantly ($P < .05$) higher than in sodium chloride-controls.

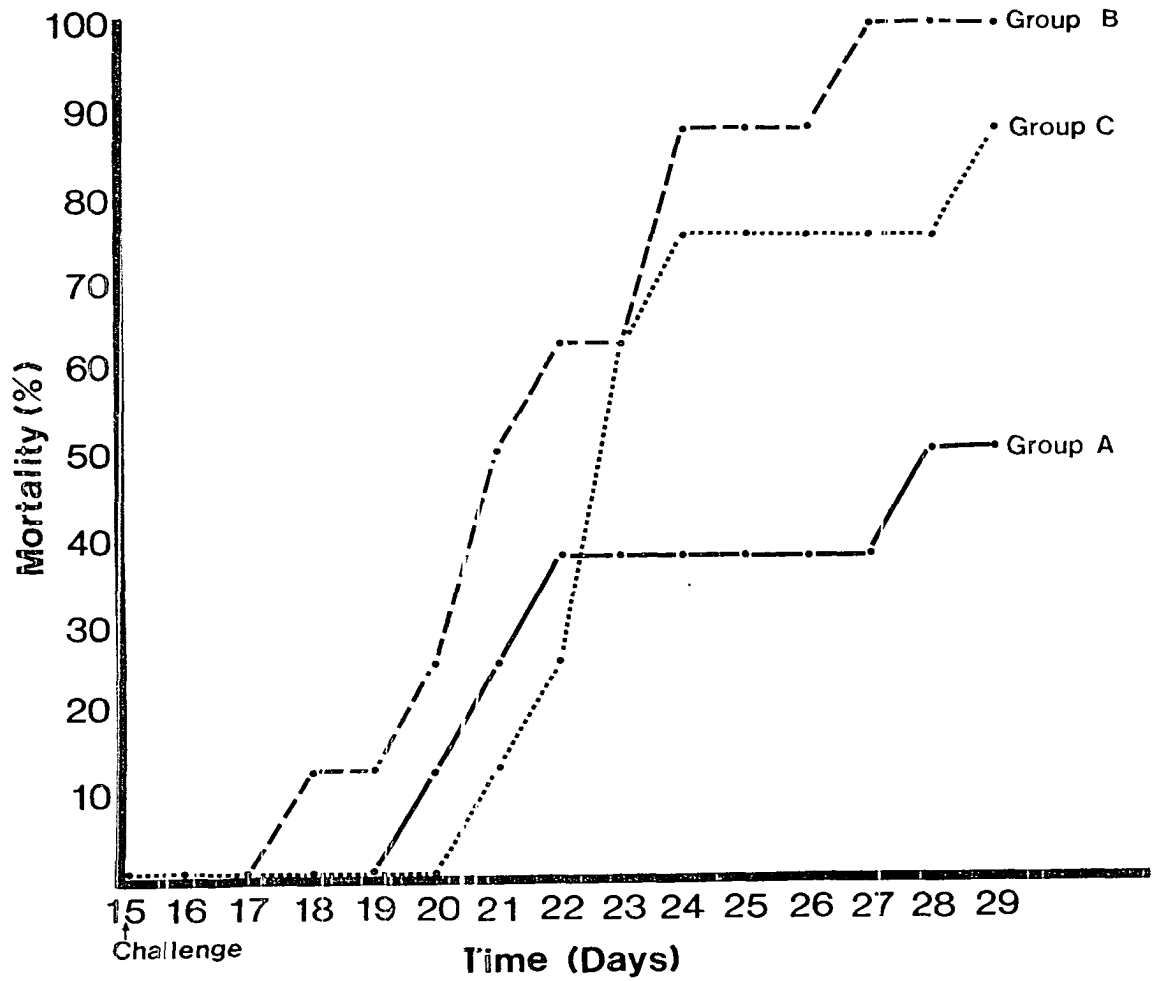
Body temperatures

While mean body temperatures of lead chloride-exposed groups were not significantly different from that of group A (sodium chloride-control) prior to Salmonella challenge, the mean body temperature of group B (2 weeks lead chloride) was significantly ($P < .05$) higher than that of group A during the post-challenge period.

Prior to challenge, daily mean body temperatures of all groups remained normal ($38.6^{\circ}$-$40.0^{\circ}$C) with the exception of a $0.2^{\circ}$C elevation noted in group A on day 14.

Mean temperatures were above normal in all groups by the third day after Salmonella challenge but returned to normal and remained normal by 9, 11, and 9 days after challenge in groups A, B, and C (1 week lead chloride), respectively.

Figure 1. Experiment 3--Daily and cumulative mortalities in groups A (sodium chloride-control), B (2 week lead chloride), and C (1 week lead chloride) following challenge with S. choleraesuis var. Kunzendorf.


100
90
80
70
60
50
40
30
20
10
Mortality (%)
Group B
Group C
Group A
15 16 17 18 19 20 21 22 23 24 25 26 27 28 29
Challenge
Time (Days)

Mean body temperatures, prior to and following challenge, and results of statistical analysis are summarized in Table 26. Daily group means are given in the Appendix (Tables 104 and 105).

Blood lead concentrations

Highly significant ($P < .01$) elevations of mean blood lead concentrations for the entire experiment were detected in both lead chloride-exposed groups as compared to group A (sodium chloride-control).

The mean blood lead concentration prior to the beginning of lead or sodium acetate exposure was 4 ug/dl. At the time of challenge, concentrations were 1.0, 97.4, and 85.6 ug/dl in groups A, B (2 weeks lead chloride), and C (1 week lead chloride), respectively.

Mean blood lead concentrations and results of statistical analysis are given in Table 26. Daily group means are summarized in the Appendix (Table 106).

Blood ALAD activities

Highly significant ($P < .01$) decreases of mean blood ALAD activities for the whole trial were noted in both lead chloride-exposed groups as compared to group A (sodium chloride-control).

Table 26. Experiment 3--Group means[a] from 2 replicates.

| Measurement | Group[b] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| Body temperature (°C) | | | | | |
| Pre-challenge[c] | 39.6 | 39.4 | 39.4 | 0.5 | 0.8 |
| Post-challenge[d] | 39.8 | 40.3* | 39.9 | 0.5 | 0.8 |
| Blood lead concentration (ug/dl) | 5 | 89** | 67** | 30 | 60 |
| Blood ALAD activity | 124.7 | 15.5** | 21.3** | 10.7 | 19.6 |

[a]Bleedings prior to lead or sodium chloride exposure not used in calculating means.

[b]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[c]Period of lead or sodium chloride exposure prior to Salmonella challenge.

[d]Following Salmonella challenge.

*Significant difference ($P < .05$) as compared to group A.

**Highly significant difference ($P < .01$) as compared to group A.

Mean blood ALAD activity prior to lead or sodium acetate exposure was 116.5 units. Following lead chloride exposure, blood ALAD activities fell sharply to about 20% of pre-exposure levels and remained at these levels at the time of challenge. Activities tended to rise in group A (sodium chloride-control) prior to challenge.

Mean blood ALAD activities and results of statistical analysis are given in Table 26. Daily group means are summarized in the Appendix (Table 107).

Hematologic observations

No significant group differences in mean blood hemoglobin concentrations were found during the period prior to Salmonella challenge. Following challenge, the mean blood hemoglobin concentration of group C (1 week lead chloride) was significantly lower than that of group A (sodium chloride-control). Mean blood hemoglobin concentrations of all groups were below normal (10-16 g/dl) throughout the pre- and post-challenge periods. Concentrations fell throughout the pre-challenge period, rose slightly by the second day after challenge and then decreased again in all groups. Decreases during the post-challenge period were most marked in lead chloride-exposed groups.

Mean hematocrit percentages of lead and sodium chloride-exposed groups were not significantly different during either

the pre- or post-challenge period and were below normal (32-50%) in all groups during both periods.

No significant group differences in total red blood cell counts were found during either the pre- or post-challenge period. Total red blood cell counts fell below normal (5-8 x $10^6$/cubic mm) in all groups on various sampling dates during the trial.

Mean MCVs of lead and sodium chloride-exposed groups were not significantly different during either the pre- or post-challenge period. Except for slight decreases in group C (1 week lead chloride) on 2 sampling dates, mean MCVs remained within normal limits (50-68 fl) throughout the experiment.

Mean MCHCs of groups B (2 weeks lead chloride) and C (1 week lead chloride) were significantly ($P < .05$) lower than that of group A (sodium chloride-control) during the period prior to Salmonella challenge. Following challenge, the mean MCHC of group B remained significantly ($P < .05$) lower than that of group A. Gradual decreases of MCHCs were noted in all groups during the pre-challenge period, and values fell below normal (30-34 g/dl) in groups B and C during this time. Mean MCHCs rose by the second day after challenge but then fell below normal by the fourth day after challenge and continued to decrease in all groups during the remainder of the post-challenge period.

During the pre-challenge period, the mean MCH of group B (2 weeks lead chloride) was significantly ($P < .05$) lower than that of group A (sodium chloride-control). Following Salmonella challenge, mean MCHs of groups B and C (1 week lead chloride) were significantly ($P < .05$) lower than that of group A. Mean MCHs were below normal in all groups on most sampling dates throughout the experiment, but lowest values occurred in groups B and C.

Mean blood hemoglobin concentrations, hematocrit percentages, total red blood cell counts, MCVs, MCHCs, and MCHs, prior to and following challenge, are given in Tables 27 and 28. Results of statistical analyses are summarized in these tables. Daily group means are given in the Appendix (Tables 108-113).

During the pre-challenge period, significant group differences in mean total leukocyte counts were not detected. After challenge, the total leukocyte count of group C (1 week lead chloride) was significantly ($P < .05$) lower than that of group A (sodium chloride-control). Mean total leukocyte counts of groups B (2 weeks lead chloride) and C were above normal (11,000-22,000/cubic mm) on some bleeding dates prior to challenge. Counts were above normal in all groups on the second and third days after Salmonella challenge and then fell to within normal limits on the fifth day after challenge. On

Table 27. Experiment 3--Group means[a] from 2 replicates. Period of lead or sodium chloride exposure prior to Salmonella challenge.

| Measurement | Group[b] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| Blood hemoglobin concentration (g/dl) | 8.6 | 8.4 | 7.8 | 0.9 | 1.5 |
| Hematocrit (%) | 28 | 28 | 26 | 3 | 5 |
| Red blood cell count (x $10^6$)[c] | 5.0 | 5.3 | 4.8 | 0.9 | 1.6 |
| MCV (fl) | 56.6 | 54.5 | 55.0 | 3.6 | 4.5 |
| MCHC (g/dl) | 31.0 | 29.4* | 29.5* | 1.1 | 1.8 |
| MCH (pg) | 17.3 | 15.9* | 16.2 | 1.3 | 2.1 |

[a]Bleedings prior to lead or sodium chloride exposure not used in calculating means.

[b]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[c]Count in cells/cubic mm.

*Significant difference ($P < .05$) as compared to group A.

Table 28. Experiment 3--Group means from 2 replicates. Period following Salmonella challenge.

| Measurement | Group[a] | | | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| | A (control) | B | C | | |
| Blood hemoglobin concentration (g/dl) | 8.3 | 7.5 | 7.2* | 0.9 | 1.5 |
| Hematocrit (%) | 28 | 27 | 26 | 3 | 5 |
| Red blood cell count (x $10^6$)[b] | 5.1 | 5.1 | 5.0 | 0.9 | 1.6 |
| MCV (fl) | 55.6 | 53.5 | 52.1 | 3.6 | 4.5 |
| MCHC (g/dl) | 29.2 | 28.1* | 28.2 | 1.1 | 1.8 |
| MCH (pg) | 16.1 | 14.7* | 14.6* | 1.3 | 2.1 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[b]Count in cells/cubic mm.

*Significant difference ($P < .05$) as compared to group A.

the seventh and ninth days post-challenge, counts were very high in group A but remained normal to slightly above normal in groups B and C.

No significant group differences in mean total segmented neutrophil counts were found during either the pre- or post-challenge period. Prior to challenge, total segmented neutrophil counts were above normal (3,200-10,000/cubic mm) in all groups on several sampling dates. Counts were above normal in all groups by 2 days post-challenge, but then fell and reached their lowest points on the fifth day after Salmonella challenge. While mean segmented neutrophil counts of groups B (2 weeks lead chloride) and C (1 week lead chloride) remained very low on the seventh and ninth days after Salmonella challenge, the count had risen and was above normal by the ninth day post-challenge in group A (sodium chloride-control).

Mean total band neutrophil counts of lead and sodium chloride-exposed groups were not significantly different during either the pre- or post-challenge period. Prior to challenge, total band neutrophil counts were above normal (0-800/cubic mm) in all groups on certain bleeding dates. Counts were above normal in group A (sodium chloride-control) by 2 days after challenge and in groups B (2 weeks lead chloride) and C (1 week lead chloride) by 3 days after challenge. While total band neutrophil counts of group A

remained stable on the fifth, seventh, and ninth days after Salmonella challenge, counts continued to rise in groups B and C on these same days.

Prior to Salmonella challenge, no significant group differences in mean total lymphocyte counts were detected, but mean counts of groups B (2 weeks lead chloride) and C (1 week lead chloride) were significantly ($P < .05$) lower than that of group A (sodium chloride-control) during the post-challenge period. Total lymphocytes counts remained within normal limits (4,500-11,000/cubic mm) in all groups throughout the experiment. While total counts in groups B and C decreased after challenge, they remained stable in group A.

Mean total eosinophil counts of lead and sodium chloride-exposed groups were not significantly different during either the pre- or post-challenge period. During the pre-challenge period, total eosinophil counts remained normal (50-2,000/cubic mm). Total counts fell below normal in groups B (2 weeks lead chloride) and C (1 week lead chloride) by the third day after challenge. On most sampling dates of the post-challenge period, mean total eosinophil counts of groups B and C were much lower than that of group A (sodium chloride-control).

No group differences in mean total monocyte counts were noted during either the pre- or post-challenge period. Total monocyte counts of all groups rose after challenge.

Mean plasma protein:fibrinogen ratios of lead and sodium chloride-exposed groups were not significantly different during either the pre- or post-challenge period.

Mean total leukocyte, segmented neutrophil, band neutrophil, lymphocyte, eosinophil, and monocyte counts and mean plasma protein:fibrinogen ratios, prior to and following challenge, are summarized in Tables 29 and 30. Results of statistical analyses are also presented in these tables. Daily group means are given in the Appendix (Tables 114-120). Daily mean total leukocyte, segmented neutrophil, and band neutrophil counts are diagramed in Figures 2-4.

Agarose film electrophoresis

Electrophoretic separation of serum protein fractions was not complete enough to allow separation of $alpha_1$ and $alpha_2$ or $beta_1$ and $beta_2$ globulin peaks. Therefore, the alpha and beta globulin fractions were each measured as 1 fraction.

No significant group differences in mean total serum protein or albumin concentrations were found during either the pre- or post-challenge period. These concentrations were highest in all groups on the second day after challenge and then decreased during the rest of the post-challenge period.

The mean serum alpha globulin concentration of group B (2 weeks lead chloride) was significantly ($P < .05$) lower than that of group A (sodium chloride-control) during the pre-challenge period, and following Salmonella challenge, mean

Table 29. Experiment 3--Group means[a] from 2 replicates. Period of lead or sodium chloride exposure prior to *Salmonella* challenge.

| Measurement | Group[b] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| Total counts[c] | | | | | |
| Leukocyte | 19,634 | 21,669 | 19,128 | 7,534 | 13,008 |
| Segmented neutrophil | 10,882 | 11,212 | 10,374 | 7,033 | 11,848 |
| Band neutrophil | 484 | 452 | 215 | 4,702 | 8,259 |
| Lymphocyte | 8,118 | 9,448 | 8,280 | 2,639 | 4,436 |
| Eosinophil | 234 | 438 | 208 | 441 | 786 |
| Monocyte | 4 | 82 | 26 | 207 | 330 |
| Plasma protein: fibrinogen ratio | 25 | 34 | 28 | 10 | 17 |

[a]Bleedings prior to lead or sodium chloride exposure not used in calculating means.

[b]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[c]Cells/cubic mm of blood.

Table 30. Experiment 3--Group means from 2 replicates. Period following Salmonella challenge.

| Measurement | Group[a] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| Total counts[b] | | | | | |
| Leukocyte | 28,583 | 23,325 | 20,973* | 7,534 | 13,008 |
| Segmented neutrophil | 9,616 | 5,909 | 5,607 | 7,033 | 11,848 |
| Band neutrophil | 8,128 | 10,474 | 7,385 | 4,702 | 8,259 |
| Lymphocyte | 10,372 | 6,601* | 7,713* | 2,639 | 4,436 |
| Eosinophil | 281 | 98 | 175 | 441 | 786 |
| Monocyte | 281 | 182 | 72 | 210 | 330 |
| Plasma protein: fibrinogen ratio | 21 | 26 | 24 | 10 | 17 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[b]Cell/cubic mm of blood.

*Significant difference ($P < .05$) as compared to group A.

Figure 2. Experiment 3--Mean total leukocyte (WBC) counts of groups A (sodium chloride-control), B (2 weeks lead chloride), and C (1 week lead chloride) prior to and following challenge with S. choleraesuis var. Kunzendorf.

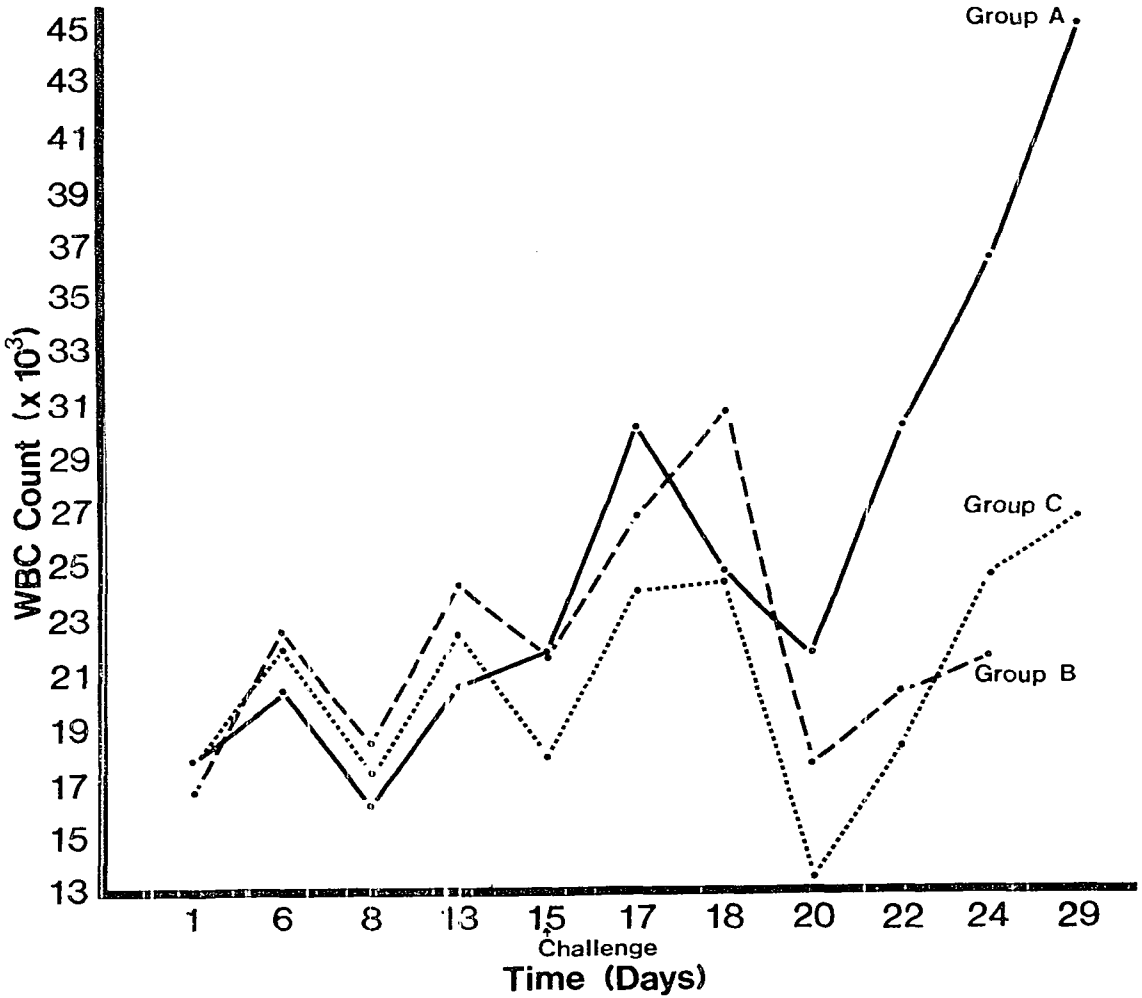

Group A
Group C
Group B
WBC Count (x $10^3$)
45
43
41
39
37
35
33
31
29
27
25
23
21
19
17
15
13
1
6
8
13
15
17
18
20
22
24
29
Challenge
Time (Days)

Figure 3. Experiment 3--Mean total segmented neutrophil counts of groups A (sodium chloride-control), B (2 weeks lead chloride), and C (1 week lead chloride) prior to and following challenge with S. choleraesuis var. Kunzendorf.

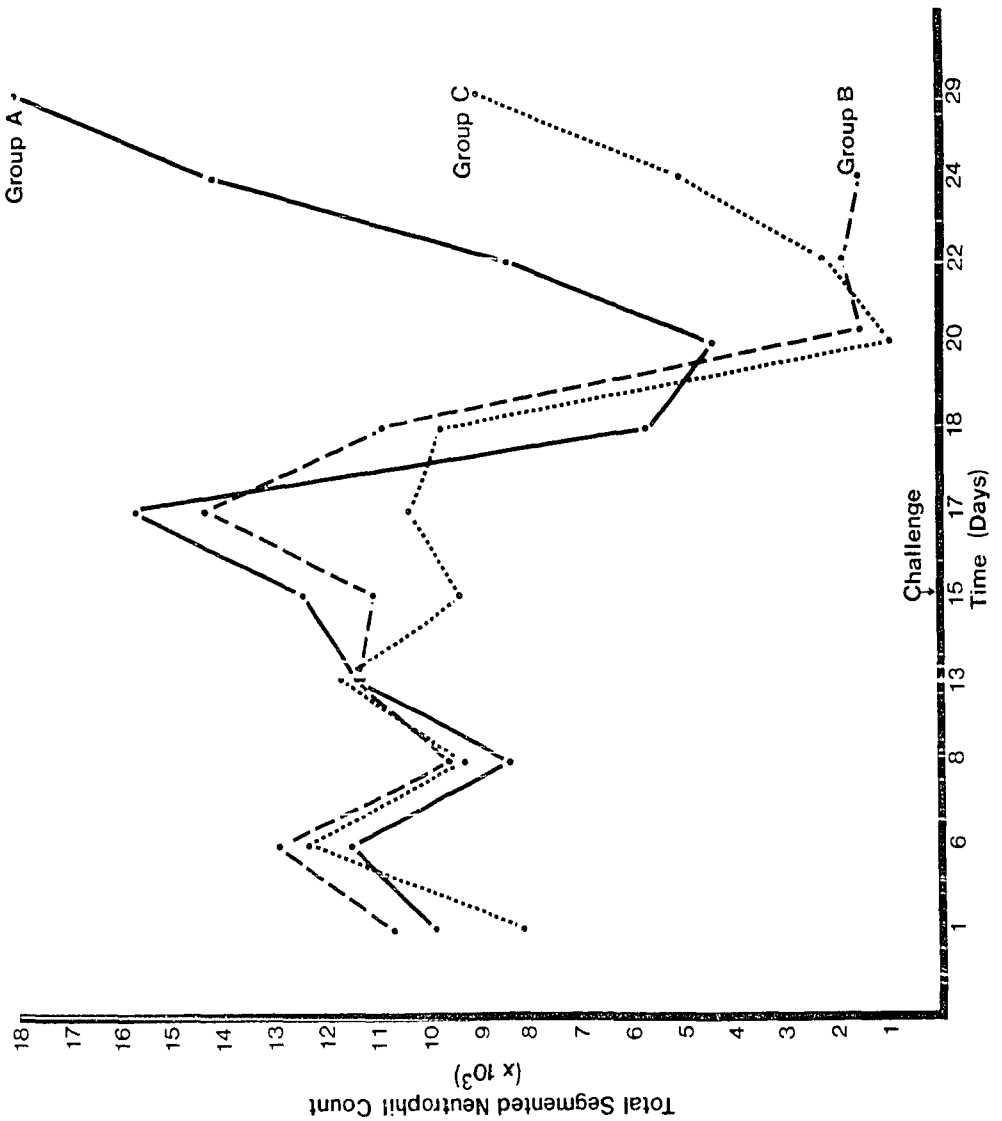
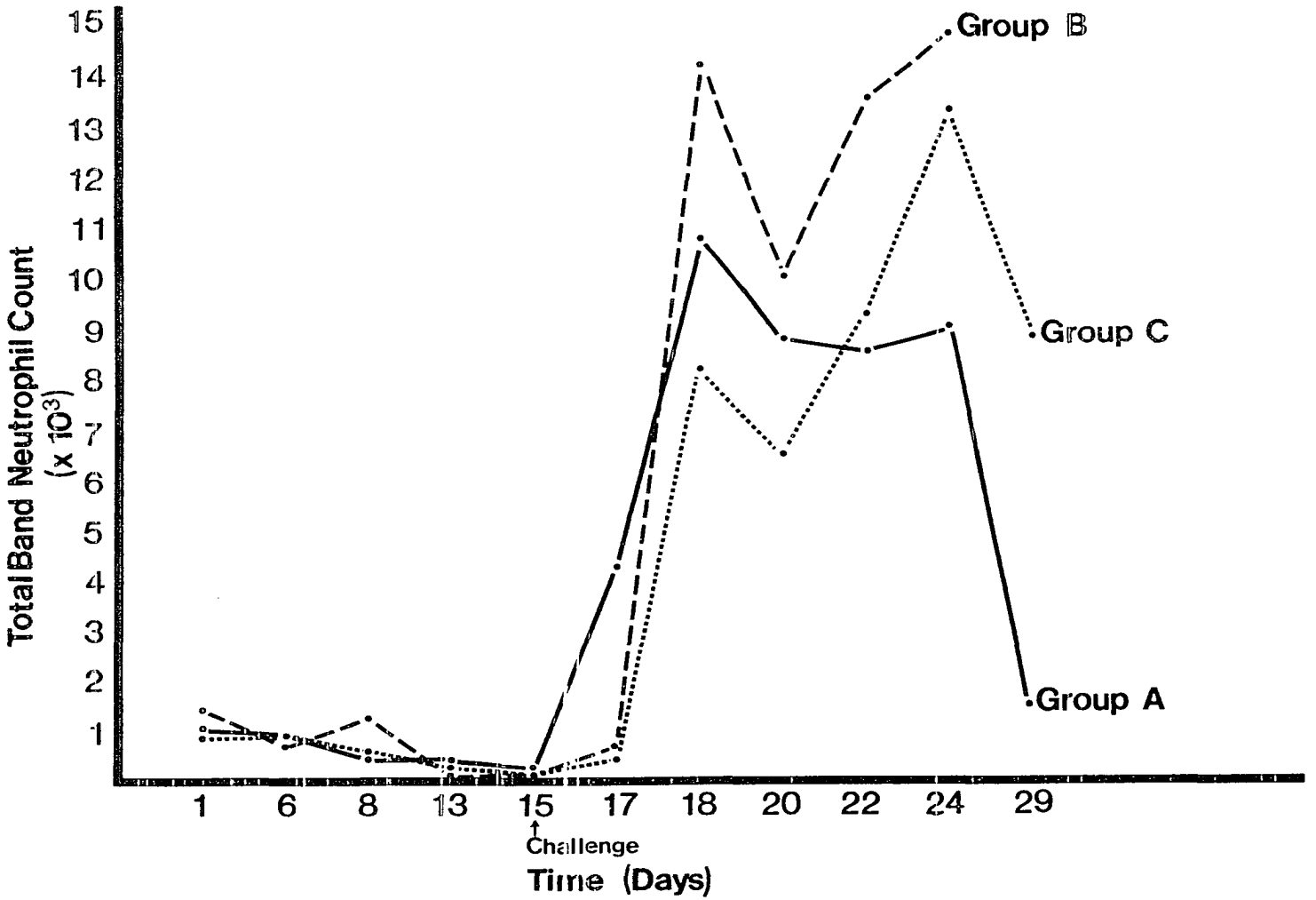

Group A
Group C
Group B
Challenge
Time (Days)
Total Segmented Neutrophil Count
(x 10³)

Figure 4. Experiment 3--Mean total band neutrophil counts of groups A (sodium chloride-control), B (2 weeks lead chloride), and C (1 week lead chloride) prior to and following challenge with *S. choleraesuis* var. *Kunzendorf*.


Group B
Group C
Group A
Total Band Neutrophil Count
(x 10³)
15
14
13
12
11
10
9
8
7
6
5
4
3
2
1
1
6
8
13
15
17
18
20
22
24
29
Challenge
Time (Days)

serum alpha globulin concentrations of both lead chloride-exposed groups were significantly ($P < .05$) lower than that of group A. Alpha globulin concentrations of all groups increased after challenge, but this increase was more marked in group A.

Mean serum beta and gamma globulin concentrations of lead and sodium chloride-exposed groups were not significantly different during either the pre- or post-challenge period. Beta and gamma globulin concentrations of all groups rose after Salmonella challenge.

The mean serum A/G ratio of group B (2 weeks lead chloride) was significantly ($P < .05$) higher than that of group A (sodium chloride-control) during the pre-challenge period. After Salmonella challenge, the mean serum A/G ratios of both lead chloride-exposed groups were significantly ($P < .05$) higher than that of group A.

Mean total serum protein, albumin, alpha globulin, beta globulin, and gamma globulin concentrations and mean albumin: globulin ratios, prior to and following Salmonella challenge, are summarized in Tables 31 and 32. Results of statistical analyses are also given in these tables. Daily group means are presented in the Appendix (Tables 121-126).

Table 31. Experiment 3--Group means[a] from 3 replicates. Period of lead or sodium chloride exposure prior to *Salmonella* challenge.

| Measurement | Group[b] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| *Serum concentrations*[c] | | | | | |
| Total protein | 4.6 | 4.4 | 4.4 | 0.6 | 0.9 |
| Albumin | 1.43 | 1.49 | 1.46 | 0.30 | 0.52 |
| *Globulins* | | | | | |
| Alpha | 1.64 | 1.41* | 1.53 | 0.20 | 0.32 |
| Beta | 0.76 | 0.76 | 0.76 | 0.14 | 0.23 |
| Gamma | 0.76 | 0.73 | 0.70 | 0.27 | 0.48 |
| Albumin:globulin ratio | 0.46 | 0.51* | 0.49 | 0.05 | 0.08 |

[a]Bleedings prior to lead or sodium acetate exposure not used in calculating means.

[b]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[c]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group A.

Table 32. Experiment 3--Group means from 2 replicates. Period following *Salmonella* challenge.

| Measurement | Group[a] A (control) | B | C | L.S.D. .05 | L.S.D. .01 |
|---|---|---|---|---|---|
| Serum concentrations[b] | | | | | |
| Total protein | 5.3 | 4.9 | 4.7 | 0.6 | 0.9 |
| Albumin | 1.44 | 1.45 | 1.45 | 0.30 | 0.52 |
| Globulin | | | | | |
| Alpha | 2.10 | 1.62** | 1.60** | 0.20 | 0.32 |
| Beta | 0.86 | 0.82 | 0.79 | 0.14 | 0.23 |
| Gamma | 0.89 | 0.93 | 0.84 | 0.27 | 0.48 |
| Albumin:globulin ratio | 0.37 | 0.43* | 0.45** | 0.05 | 0.08 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[b]Concentrations in g/dl.

*Significant difference ($P < .05$) as compared to group A.

**Highly significant difference ($P < .01$) as compared to group A.

Serum agglutinin titrations

Serum samples obtained prior to assignment of pigs to groups were negative for S. choleraesuis var. Kunzendorf agglutinins.

Salmonella choleraesuis var. Kunzendorf somatic and flagellar agglutinin titers of lead and sodium acetate-exposed groups following Salmonella challenge were not significantly different.

Daily mean somatic and flagellar agglutinin titers on a day prior to and on several days following challenge are given in Tables 33 and 34.

Salmonella isolation from tissues and rectal swabs

Salmonella was not isolated from rectal swabs obtained prior to assignment of pigs to groups.

Salmonella was isolated from 15 of 16 lead chloride-exposed pigs and 6 of 8 sodium chloride-control pigs. Isolates which were serotyped were all determined to be S. choleraesuis var. Kunzendorf.

Results of attempted Salmonella isolation from selected tissues are given in Table 35.

Tissue lead concentrations

Mean liver lead concentrations were 4.23, 10.78, and 10.33 ppm and mean kidney lead concentrations were 2.80, 9.23, and 7.83 ppm in groups A, B, and C, respectively.

Table 33. Experiment 3--Geometric means of S. choleraesuis var. Kunzendorf somatic agglutinin titers on a daily basis.

| Day[b] | Group[a] | | |
|---|---|---|---|
| | A (control) | B | C |
| 13 | 0 | 0 | 0 |
| 17 | 0 | 0 | 0 |
| 20 | 8 | 2 | 2 |
| 22 | 52 | 40 | 40 |
| 24 | 60 | 86 | 65 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[b]Pigs were challenged with Salmonella on day 15.

Table 34. Experiment 3--Geometric means of S. choleraesuis var. Kunzendorf flagellar agglutinin titers on a daily basis.

| Day[b] | Group[a] | | |
|---|---|---|---|
| | A (control) | B | C |
| 13 | 0 | 0 | 0 |
| 17 | 0 | 0 | 0 |
| 20 | 0 | 0 | 0 |
| 22 | 22 | 20 | 25 |
| 24 | 60 | 120 | 70 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

[b]Pigs were challenged with Salmonella on day 15.

Table 35. Experiment 3--Positive *Salmonella* isolates from selected tissues/number of pigs in replicate or group.

| Group[a] | Tissue | | |
|---|---|---|---|
| | Liver | Mesenteric lymph node | Ileum |
| A (control) | | | |
| Rep 1 | 3/4 | 3/4 | 3/4 |
| Rep 2 | 3/4 | 3/4 | 3/4 |
| Total | 6/8 | 6/8 | 6/8 |
| B | | | |
| Rep 1 | 4/4 | 4/4 | 4/4 |
| Rep 2 | 4/4 | 4/4 | 3/4 |
| Total | 8/8 | 8/8 | 7/8 |
| C | | | |
| Rep 1 | 3/4 | 3/4 | 3/4 |
| Rep 2 | 4/4 | 4/4 | 3/4 |
| Total | 7/8 | 7/8 | 6/8 |

[a]Groups B and C exposed to lead chloride for 2 and 1 week, respectively.

Macroscopic lesions

Spleens of 50% of the pigs (10 of 16 lead chloride-exposed; 2 of 8 sodium chloride-control) were enlarged 2-4 times and dark red.

Consolidated areas were found in lungs of 6 pigs (4 of 16 lead chloride-exposed; 2 of 8 sodium chloride-control). In most pigs, scattered lobules were consolidated while in

1 pig a 4 x 2 x 2 cm area was involved. Anterior and middle lobes were most frequently altered; however, the diaphragmatic lobe was also involved in 2 pigs.

A crusty to mucoid, yellow-white material covered the mucosal surface of portions of the intestinal tract in most pigs (14 of 16 lead chloride-exposed; 6 of 8 sodium chloride-control). This material was found most consistently in the cecum and colon but frequently also lined portions of the caudal ileum. In some pigs, the material was present in elevated foci on the mucosal surface (button ulcers). Intestinal lesions were numerically rated on the following basis:

1--Necrosis not grossly evident.

2--Focal necrosis only.

3--Diffuse necrosis of cecum and colon with slight involvement of ileum (i.e. 60 cm or less).

4--Diffuse necrosis of cecum and colon with major involvement of ileum (i.e. greater than 60 cm).

The average score of lead chloride-exposed pigs was 2.6 while that of sodium chloride-control pigs was 2.0.

The mucosa of other portions of the gastrointestinal tract was reddened in many pigs.

In 1-2 pigs from each group, the peritoneum was covered with a white, milky material, and abdominal organs tended to adhere to each other.

Microscopic lesions

Acute, multifocal necrotic hepatitis characterized by coagulative necrosis with no definite lobular distribution, moderate hyperplasia of Kupffer cells, increased neutrophil numbers in sinusoids, and accumulations of mononuclear inflammatory cells in portal areas was detected in 9 pigs (4 of 16 lead chloride-exposed; 5 of 8 sodium chloride-control). Neutrophils, lymphoreticular cells, or large numbers of erythrocytes were present in various necrotic foci.

Lymphoid depletion was evident in all spleens. Congestion and extramedullary hematopoiesis were also found in spleens of many lead or sodium chloride-exposed pigs. Splenitis characterized by focal to diffuse neutrophil accumulations was detected in a small number of pigs.

An acute diffuse interstitial pneumonia characterized by accumulations of macrophages and neutrophils in alveolar walls was evident in most pigs. In a few pigs, alveoli and bronchioles contained: abundant fibrin, neutrophils, macrophages, and necrotic debris.

Lymphoid depletion was detected in all mesenteric lymph nodes examined. Lymphadenitis characterized by micro-abscesses, edema, and multifocal coagulative necrosis was found in 1 lymph node.

Acute to subacute colitis, typhlitis, and ileitis characterized by focal to diffuse coagulative necrosis, edema,

and accumulations of neutrophils, eosinophils, macrophages, and lymphocytes in the mucosa and/or submucosa were detected in pigs from all groups. Fibroplasia was evident in the submucosa of the pigs which died late in the experiment.

Congestion and a slight increase in neutrophil numbers on the mucosal surface were the only lesions observed in the jejunum and duodenum.

Focal collections of neutrophils and fibrin were present on serosal surfaces of various sections of intestine.

No microscopic lesions were observed in kidneys.

# DISCUSSION

## Experiment 1

This experiment was performed to detect physiopathologic effects of lead in the pig, including levels of lead exposure which would and would not cause clinical toxicosis. Such a study was necessary before subclinical levels of lead exposure could be chosen for use in experiments 2 and 3.

This research demonstrated that pigs are extremely tolerant of lead. Link and Pensinger (1966) reported mean blood lead concentrations of 120 ug/dl at the time of death in lead-poisoned pigs, but in the present research, several i.p. lead acetate-exposed pigs had blood lead concentrations exceeding 120 ug/dl at various times, and 1 pig from group 9 (16 mg/kg lead acetate i.p.) had an extremely high blood lead concentration (up to 14,300 ug/dl), but these pigs did not die of lead toxicosis.

The first report of lead toxicosis in swine was by Bywater (1937) who observed anorexia, incoordination, hindlimb paralysis, abortions, passing of black, bloody feces, purple-blue discoloration of skin, and death (over 1 month after the suspected exposure date). Lead concentration in "ingesta and organs" was 114 ppm. The possible role of infectious disease was not thoroughly investigated in this report.

Link and Pensinger (1966) reported on experimentally-induced lead toxicosis in swine. Oral lead acetate exposure rates of either 11 or 66 mg elemental lead per kg body weight did not cause acute lead toxicosis. Pigs developed mild diarrhea after 3 days of lead exposure but this subsided after 7 to 11 days. Other clinical signs observed during a 16-week lead-exposure period were inappetance, listlessness, muscle tremors, incoordination, increased respiratory rates, enlarged carpal joints, and mild clonic seizures. Five of 8 pigs died within 90 days after initiation of lead exposure.

In the current research, the coughing observed in 1 orally lead acetate-exposed and 1 orally sodium acetate-exposed pig was probably due to either a transient respiratory infection or inhalation of the fluid which was administered.

Gauntness and rough coats were noted in orally lead acetate-exposed pigs but not in orally sodium acetate-exposed controls and might have been a manifestation of mild intoxication. If this were the case, the pigs adapted, became normal, and remained normal despite higher blood lead burdens later in the experiment.

The significance of the clinical signs observed in i.p. lead acetate-exposed pigs is clouded by the fact that i.p. injections of lead acetate caused a fibrinous to granulomatous peritonitis. Most of the signs observed could be attributed to chronic peritonitis with adhesions. Lateral

nystagmus and rhythmic jerking of the head and forelimbs in 1 pig prior to death may have been neurological manifestations of lead intoxication but could also have been caused by electrolyte and/or acid-base imbalances resulting from intestinal obstruction.

It appears that the alterations of mean blood hemoglobin concentrations, hematocrit percentages, MCHCs, and MCHs detected in experiment 1 are related to lead dosage and to blood lead concentrations. These values were significantly decreased in i.p. lead acetate-exposed groups, which had the highest blood lead concentrations, but were not significantly decreased in orally lead acetate-exposed groups, which had blood lead concentrations uniformly lower than those of i.p. lead acetate-exposed groups. Furthermore, mean blood hemoglobin concentrations and hematocrit percentages fell below accepted normal limits and were significantly decreased only in surviving pigs from groups 8 (8 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.). These same pigs had the highest blood lead concentrations of any in the experiment. Lowered blood hemoglobin concentrations were also reported in lead-poisoned pigs by Link and Pensinger (1966).

The significant decrease in mean MCV which occurred in group 8 (8 mg/kg lead acetate i.p.) may be related to lead treatment, but since a significant decrease in MCV did not occur in group 9 (16 mg/kg lead acetate i.p.), the

relationship of this alteration to lead dosage and blood lead concentration is not clearly established.

There are many reports concerning the influence of lead on red blood cells. Possible lead-induced alterations of erythrocytes are: depression of heme synthesis, alterations of osmotic fragility, and inducement of basophilic stippling.

It has been suggested that lead interferes with the activities of delta-aminolevulinic acid (ALA) synthetase (Goldberg et al., 1956), delta-aminolevulinic acid (ALA) dehydratase (Gibson et al., 1955), and heme synthetase (Goldberg et al., 1956). All of these enzymes catalyze reactions in the normal scheme of heme synthesis as noted in Figure 5.

The current investigation revealed that blood ALAD activity in the pig is rapidly and drastically reduced by lead exposure. Gibson et al. (1955) demonstrated in vitro inhibition of ALAD activity by lead, and lead-induced inhibition of ALAD activity in vivo has subsequently been reported in humans (Nakao et al., 1968), cattle (Green et al., 1973), rats (Millar et al., 1970), and hamsters (Mankus, 1975). Gibson and Goldberg (1970) concluded that since glutathione could partially reverse lead-induced ALAD inhibition, the mechanism of such inhibition was probably through inactivation of the enzyme's sulfhydryl groups.

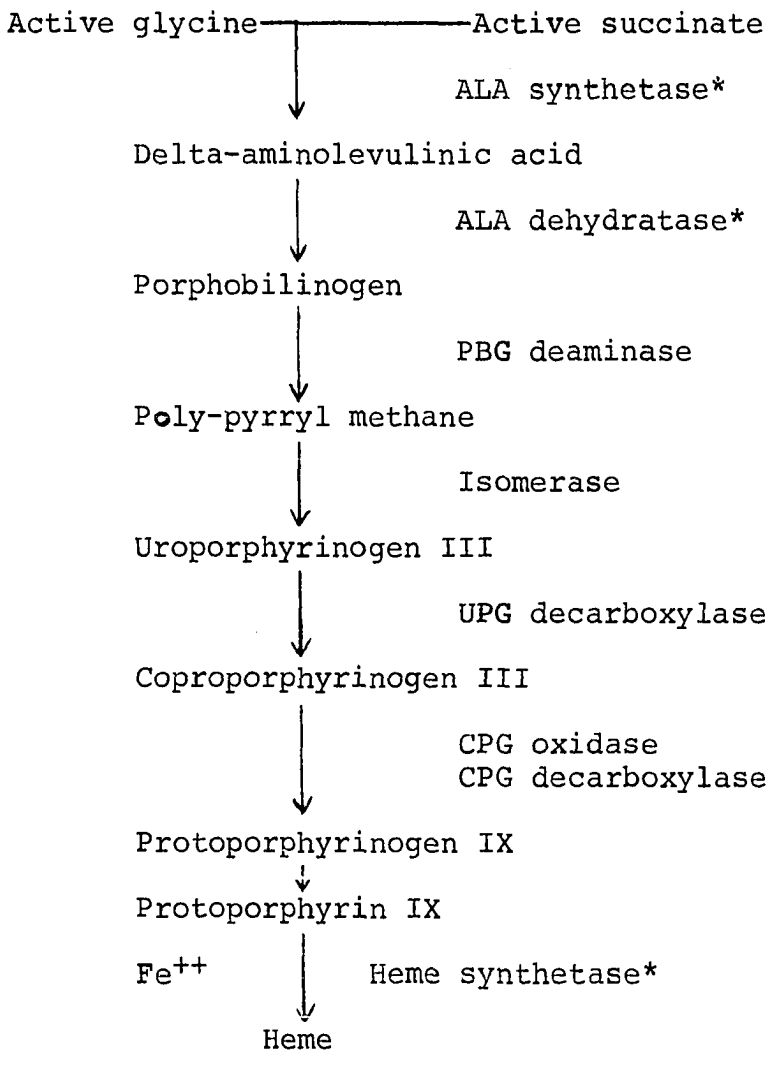



Figure 5. Schematic outline of heme synthesis (Griggs, 1964). Asterisks indicate enzymes most markedly inhibited by lead.

Although lead clearly inhibits enzymes involved in catalyzing heme synthesis, the ultimate effect of this inhibition on blood hemoglobin concentrations and hematocrit percentages is not clear. Sakurai et al. (1974) found no changes in hematocrits, red blood cell counts, or blood hemoglobin

concentrations in chronically lead-poisoned humans with blood lead concentrations up to 50 ug/dl, and Mankus (1975) detected only minor decreases in blood hemoglobin concentrations and hematocrit percentages in lead-exposed hamsters with marked decreases in blood ALAD activity. Haeger-Aronsen et al. (1971) estimated that before significant lead-induced impairment of heme synthesis could occur, blood ALAD activity would have to fall to 1/3 of normal.

In the present research, it was apparent that blood ALAD activities may remain below 30% of their original levels for nearly 12 weeks without significant detectable decreases in hemoglobin synthesis resulting. Bush et al. (1955) found the average potential life-span of swine erythrocytes to be 86 $\pm$ 11.5 days. This means that almost all erythrocytes would have been replaced during the 90 day period of the experiment, and, yet, mean blood hemoglobin concentrations and MCHCs remained within normal limits in groups exposed to lead acetate orally (groups in which blood ALAD had fallen to approximately 30% or less of original levels after 1 week of lead exposure).

Although measured laboratory values did not clearly demonstrate a more profound inhibition of ALAD activity in i.p. than in orally lead acetate-exposed groups, the fact that significant reductions in blood hemoglobin concentrations, MCHCs, and MCHs were detected in i.p. lead acetate-exposed

groups infers that lead-sensitive enzymes of the heme synthesis pathway were more markedly inhibited in these groups.

The gradual fall of blood ALAD activities in sodium acetate-exposed groups may have been caused by very low-level lead exposure via contact with washings from adjacent pens of lead-exposed pigs or by a gradual natural fall in activity as pigs became older and erythrogenesis became less intense. Niyo (1975) reported that blood ALAD activities of young pigs rose until they reached 3-4 weeks of age and then fell gradually.

Lead-induced changes in erythrocyte osmotic fragility were not detected in experiment 1 and probably played no part in changes of erythrocyte values. Harris and Greenberg (1954) suggested that such changes were due to shrunken erythrocytes, resulting from lead-induced potassium loss, having more capacity to swell before lysis. Erythrocytes of both lead acetate-exposed and sodium acetate-control pigs were more resistant to osmotic changes than has been previously reported in either baby or 7-month-old pigs (Hudson, 1955).

The fact that basophilic stippling of erythrocytes was found in lead acetate-exposed but not in sodium acetate-control pigs indicates that the phenomenon was related to lead acetate exposure. This was an extremely rare finding in experiment 1. Link and Pensinger (1966) found basophilic

stippling in erythrocytes of lead-poisoned pigs only after they became moribund, but in the present study, basophilic stippling was observed in erythrocytes of lead acetate-exposed pigs with no clinical signs.

Jensen et al. (1965) demonstrated, in ultrastructural studies, that basophilic stippling consisted of ribosomal aggregates. These workers suggested that the aggregates probably formed as a blood smear was dried and that slow drying predisposed to stippling. Furthermore, it was felt that rather than being caused by direct ribosomal alterations, basophilic stippling may be a manifestation of less obvious chemical alterations of other cellular constituents.

Since many of the significant alterations in mean total leukocyte and differential counts observed in this investigation did not occur consistently in groups exposed to highest lead levels, these alterations are probably not related to the exposure. One exception is group 9 (16 mg/kg lead acetate i.p.) where leukocytosis with absolute neutrophilia and a left shift were detected. Considering the extensive peritonitis caused by i.p. lead acetate injections, these findings are less surprising than is the fact that similar changes were not noted in other i.p. lead acetate-exposed groups. It is apparent that inflammation was severe enough to cause detectable effects on circulating white blood cells only in group 9.

Reports concerning the effect of lead exposure on leukocytes are rare. Bell et al. (1925) found a tendency toward leukocytosis with relative lymphocytosis and eosinophilia in human cancer patients treated therapeutically with colloidal lead. Sharma (1971) reported significant increases in band neutrophil and monocyte counts and significant decreases in eosinophil counts in lead-exposed sheep. Significant increases in mean absolute band neutrophil and eosinophil counts were detected in groups 2 (16 mg/kg lead acetate orally) and 7 (4 mg/kg lead acetate i.p.), respectively, but not in most other groups with higher mean blood lead levels. The significance of these findings is, therefore, questionable.

It must be concluded that except for direct irritation, the lead-exposure levels in this study did not significantly affect total leukocyte or differential counts.

A significantly lower mean plasma protein:fibrinogen ratio was found only in group 9 (16 mg/kg lead acetate i.p.). This indicates that there was an increased plasma concentration of fibrinogen (Schalm, 1970) which was probably a response to inflammation of the peritoneal cavity (Schalm et al., 1970). Such a finding correlates well with the previously mentioned leukocyte alterations in this group.

The only serum protein alterations which appear to be related to lead exposure in the present study are decreases

in serum albumin concentrations and in albumin:globulin ratios and increases in serum gamma globulin concentrations. It is possible that mild lead-induced liver injury inhibited albumin synthesis or that decreased feed intake, which was not noted clinically, caused decreased serum albumin concentrations. The finding of significant increases in mean serum gamma globulin concentrations in orally lead acetate-exposed group 4 and i.p. lead acetate-exposed groups 7, 8, and 9 are not in agreement with results reported by Hemphill (1973) in lead nitrate-exposed mice. It must be recognized that 2 pigs in group 4 had chronic pleuritis and pericarditis, and that many pigs in groups 7, 8, and 9 experienced peritonitis. These inflammatory changes were not observed in sodium acetate-control pigs, and, therefore, pigs in control groups were not antigenically stimulated to the same extent as were pigs in the groups with increased serum gamma globulin concentrations.

Decreased A/G ratios were a result of decreased serum albumin concentrations coupled with increased serum gamma globulin concentrations in groups 4, 7, 8, and 9.

A significant decrease in mean serum $alpha_2$ globulin concentration was noted in group 3 (32 mg/kg lead acetate orally), but a significant increase in this same fraction was detected in groups 6 (2 mg/kg lead acetate i.p.) and 9 (16 mg/kg lead acetate i.p.). While significant increases in

mean serum $beta_1$ globulin concentrations occurred in groups 3 (32 mg/kg lead acetate orally) and 8 (8 mg/kg lead acetate i.p.), significant alterations did not occur in groups 4 (64 mg/kg lead acetate orally) or 9 (16 mg/kg lead acetate i.p.), both of which had higher mean blood lead concentrations than the former groups. Although significant decreases in mean serum $beta_2$ globulin concentration occurred in groups 3 (32 mg/kg lead acetate orally) and 4 (64 mg/kg lead acetate orally), such decreases did not occur in any i.p. lead acetate-exposed groups. In all of these cases, a definite association between the occurrence of the alteration in protein concentration and either lead exposure level or blood lead concentration is not clearly established. The relationship of such alterations to lead exposure is, therefore, questionable.

In lead-poisoned pigs, Link and Pensinger (1966) found highest tissue lead concentrations to be in the femur followed by liver, kidney, muscle, and brain, in that order. The current trial confirmed that highest tissue lead concentrations are found in bone. High lead concentrations were also found in liver and kidney, but in certain groups, spleen, lung, bone marrow, or lymph node exceeded the latter organs in lead concentration.

Because of its intimate contact with bone, it is not surprising that high lead concentrations were found in the bone marrow, and such a finding indicates that large amounts

of lead are present at the main site of erythrogenesis and that there is ample opportunity for a lead-mediated effect on this process.

High lead concentrations in the spleen or lung are more difficult to explain. Lead is rapidly associated with erythrocytes following absorption (Barltrop and Smith, 1971), and normal destruction of erythrocytes takes place in the spleen. It seems that such erythrocyte destruction would result in lead being deposited in the spleen. The cause of high lung lead concentrations might be the result of accidental entrance of orally administered lead acetate solutions into the lungs; however, lung lead concentrations were also elevated in i.p. lead acetate-exposed pigs. The reason for high lung lead concentrations remains a mystery.

High concentrations of lead found in lymph nodes of i.p. lead acetate-exposed pigs can be explained by the fact that some of the lymph nodes used for analysis drained areas where lead acetate was injected. Of more importance is the fact that low lead concentrations were detected in lymph nodes of pigs exposed to lead acetate orally. This suggests that if lead could affect antibody synthesis in lymphoid tissues other than the spleen, it would have to be active at a low concentration.

This investigation confirmed that little lead is concentrated in skeletal muscle or brain.

Link and Pensinger (1966) reported that macroscopic lesions of lead-poisoned pigs included thickening of the mucosa of the stomach and intestines, a few petechiae on the heart, and congestion of the liver. These changes were not found in the current work.

The pleural and pericardial adhesions noted in pigs from group 4 (64 mg/kg lead acetate orally) were possibly due to a serositis of infectious etiology. No cultures were performed to verify this. The macroscopic lesions observed in i.p. lead acetate-exposed groups were clearly caused by a direct irritant action of lead acetate solutions. Such an effect has previously been described (Truscott, 1970).

Hsu et al. (1973) reported abundant intranuclear inclusion bodies in liver cord cells and renal tubular epithelial cells of pigs exposed to lead acetate orally for up to 13 weeks. Such inclusions were found in a few pigs in the present research. Goyer et al. (1970b) suggested that the intranuclear inclusion bodies observed in lead-poisoning consisted of a lead-protein complex. Ultrastructurally, such inclusions had a dense central core and a fibrillar outer zone. These workers theorized that such inclusions served as a depot for intracellular lead. Goyer et al. (1970a) stated that concentration of lead in intranuclear inclusions might be a defense mechanism which reduces lead concentrations in lead-sensitive organelles such as mitochondria.

It is apparent that lead acetate can incite a granulomatous reaction and that cells ingesting lead acetate may carry the substance to lymph nodes. The granulomatous masses in the abdominal cavity and the granulomatous foci in lymph nodes of i.p. lead acetate-exposed pigs contained amorphous material which was partially mineralized. This material probably represents deposits of lead salts. Selye et al. (1962) reported that lead acetate can induce calcinosis. Whether the mineralization observed in this experiment was caused by a specific effect of lead acetate or was due to a non-specific reaction to tissue injury, is not clear.

The slight interstitial thickening observed in lungs of pigs exposed to lead acetate orally might have been caused by inhalation of lead acetate solutions during the process of administration.

Link and Pensinger (1966) found degeneration of renal tubules in lead-exposed pigs, but such a change was not detected in experiment 1.

## Experiments 2 and 3

The significantly higher mortality rate observed in lead chloride-exposed pigs challenged with S. choleraesuis var. Kunzendorf in experiment 3 paralleled observations of Hemphill (1973) in lead nitrate-exposed mice challenged with

S. typhimurium. Mankus (1975), however, found no decrease in resistance to S. typhimurium in lead chloride-exposed hamsters. Furthermore, decreased resistance to S. choleraesuis var. Kunzendorf was not detected in lead acetate-exposed pigs in experiment 2.

Lead exposure levels in experiments 2 and 3 were adequate to produce significant elevations of blood lead concentrations without causing overt toxicosis. It must be noted that reduced appetites occurring in group B (2 weeks lead chloride) of experiment 3 during the lead-exposure period might have been due to mild toxicosis. At the time of challenge, blood lead concentrations of lead-exposed pigs were approximately twice as high in experiment 3 as in experiment 2. Liver and kidney lead concentrations were also higher in experiment 3. If lead exposure played a role in the increased mortality rates of experiment 3, the fact that such a phenomenon did not occur in experiment 2 might be explained by lower lead concentrations in blood and other tissues of lead-exposed pigs in this experiment and, therefore, less lead-induced reduction of resistance.

In both experiments, lead concentrations in blood and other tissues of lead-exposed pigs were adequate to cause a profound biological effect as demonstrated by marked reductions of blood ALAD activity. Such reductions in activity probably result from lead-induced inactivation of

the enzyme's free sulfhydryl groups. It might be inferred that lead levels in these experiments should also have been adequate to interfere with sulfhydryl groups of antibodies.

Lead-exposed pigs did not appear to be more severely ill than control pigs in either experiment 2 or 3. Although clinical signs were similar in both experiments, there were some qualitative and/or quantitative variations. Cyanosis was observed only in experiment 2 while diarrhea and dehydration were more consistently evident and more severe in experiment 3. Cyanosis is more commonly observed in the acute septicemic form of salmonellosis while diarrhea is more common in the enteric form of the disease (Barnes and Sorensen, 1975). The obvious conclusion is that organisms inoculated i.p. in experiment 2 had more of a tendency to cause septicemia while those inoculated intragastrically tended to cause enteric disease. While the latter inference is probably valid, culture results reveal that septicemia was more common in experiment 3 than in experiment 2.

The significance of increased mortality rates observed in lead chloride-exposed pigs in experiment 3 is somewhat diminished by the fact that decreases of blood hemoglobin concentrations, MCHCs, and MCHs occurred in these groups. Such decreases may have been manifestations of decreased hemoglobin synthesis resulting from a toxic effect of lead or may have been due to some intrinsic abnormality of

hemoglobin synthesis and/or erythrocyte destruction which was more evident in lead chloride-exposed groups. If the former theory is true, it might be invalid to classify the pigs as subclinically intoxicated. The role, if any, played by abnormalities in hemoglobin synthesis and/or erythrocyte destruction in decreasing resistance of lead-exposed pigs to Salmonella is not clear.

Although significant changes in total leukocyte and differential counts following Salmonella challenge in experiment 3 were limited to reductions of the total leukocyte count in group C (1 week lead chloride), and total lymphocyte counts in groups B (2 weeks lead chloride) and C, important differences in leukocyte kinetics were noted by 5-7 days post-challenge. Total leukocyte and segmented neutrophil counts of groups A, B, and C rose during the first 2-4 days after challenge but had fallen by 5 days post-challenge. These counts decreased more drastically and stayed lower in groups B (2 weeks lead chloride) and C (1 week lead chloride) than in group A (sodium chloride-control). Total band neutrophil counts of group A rose sooner than those of groups B and C following challenge but were below counts of the former groups after 17 days post-challenge.

Following i.v. administration of live, virulent S. choleraesuis var. Kunzendorf organisms or purified endotoxin from these organisms to pigs, Sherman (1972) detected

a leukopenia followed by a leukocytosis. The leukopenic phase was characterized by a decrease in absolute segmented neutrophil and lymphocyte counts. The phase of increasing leukocyte counts was characterized by a marked increase in absolute immature neutrophil counts. All changes were more marked in pigs which received purified endotoxin.

An explanation for endotoxin-induced changes of differential counts was given by Athens et al. (1961) who discovered that radiolabeled neutrophils moved from the circulating granulocyte pool (CGP) of the blood to the marginal granulocyte pool (MGP) following administration of endotoxin and that during this time granulocytopenia occurred. These workers noted that such events were followed by a phase of increasing granulocyte numbers in both the CPG and MGP caused by an influx of cells from the bone marrow pool.

The leukocyte changes in experiment 3 were compatible with the previously described endotoxin-induced changes. The fact that total leukocyte and segmented neutrophil counts fell more drastically and remained lower and that band neutrophil counts were higher in lead chloride-exposed pigs than in sodium chloride-exposed controls seems to indicate a more profound effect of endotoxin in the former lead chloride-exposed pigs.

Since a large number of previous studies have demonstrated that lead acetate can induce increased susceptibility

to endotoxin and since leukocyte alterations in experiment 3 indicate that lead chloride-exposed pigs may have been more profoundly affected by endotoxin than were sodium chloride-exposed controls, it is tempting to explain the increased mortality of lead chloride-exposed pigs on the basis of a lead chloride-induced endotoxin hypersensitivity. It must be noted, however, that the previously mentioned alterations could also be caused by increased amounts of endotoxin entering the bloodstream of lead chloride-exposed pigs as a result of lead-induced immunosuppression. Such immunosuppression might have allowed more rapid multiplication of bacteria which, in turn, produced increased endotoxin.

Significantly lower serum alpha globulin concentrations in lead-exposed groups prior to Salmonella challenge in experiment 3 and following Salmonella challenge in experiments 2 and 3 parallel observations by Hemphill (1973) in lead-exposed mice, Hoffman (1974) in lead-exposed sheep, and Soliman et al. (1970) in lead-exposed humans. In both experiments 2 and 3, serum alpha globulin ($alpha_1$ globulin in experiment 2) concentrations rose following Salmonella challenge, but the magnitude of this rise was greater in control pigs than in lead-exposed pigs. Soliman et al. (1970) attributed lower serum $alpha_1$ and $alpha_2$ globulin concentrations to lead-induced urinary protein loss. Since urine

proteins were not measured in experiments 2 and 3, this explanation can neither be accepted nor refuted.

Of greater importance than the significantly lower serum alpha globulin concentrations is the fact that despite increased mortality of lead chloride-exposed pigs in experiment 3, no significant decreases in serum gamma globulin concentrations were detected. This finding casts doubt on the importance of decreased humoral immunity in causing the reduced resistance to Salmonella challenge. This doubt is strengthened by the fact that significant decreases in either flagellar or somatic agglutinin titers of lead chloride-exposed pigs were not detected in experiment 3.

It must be emphasized that despite the fact that lead-exposure levels in experiments 2 and 3 caused a marked biological effect (decreased ALAD activity), there is no evidence that such exposure caused an alteration of humoral immunity as measured by serum agglutinin titers or serum beta and gamma globulin concentrations.

It is possible that a decreased cellular immune response caused lowered resistance to Salmonella in experiment 3. Collins (1974) concluded that while specific humoral factors probably make an important contribution during the early stages of the immune response to Salmonella, the organisms are protected from the actions of circulating serum factors once they become established in the lymphoreticular tissues of the

host. This reviewer contended that the host is able to eliminate organisms from the tissues only after a cellular immune response is established. Mackaness (1971) felt that the effectors of such a response were "activated" macrophages.

Investigations of the endotoxin-sensitizing effect of lead acetate indicate that this compound may impair phagocytosis and/or cause ultrastructural alterations in phagocytic cells. If such an action took place in lead chloride-exposed pigs in experiment 3, a reduction in cellular immune response and, therefore, a decreased resistance to Salmonella would have resulted.

The role of other lead-induced abnormalities such as hepatocyte injury cannot be ruled-out as playing a part in the decreased resistance of lead chloride-exposed pigs in experiment 3. Hepatic function tests were not performed during this trial and, therefore, the possibility of lead-induced hepatic injury cannot be evaluated.

There was no indication that lead-exposure potentiated the spread of organisms throughout the body. In experiment 3, organisms were isolated from most tissues cultured from both lead and sodium chloride-exposed pigs. In experiment 2, Salmonella was actually isolated from more sodium acetate-exposed pigs and from more tissues of sodium acetate-exposed pigs than from lead acetate-exposed pigs. The fact that organisms were more widespread in experiment 3 is probably a

function of the route of inoculation. Organisms inoculated i.p. in experiment 2 were probably quickly phagocytized by peritoneal macrophages and other R-E tissues of the body which resulted in a relatively small number of organisms surviving and multiplying. In experiment 3, organisms inoculated into the G-I tract had an excellent environment in which to multiply and then invade the rest of the body.

The fact that macroscopic lesions of spleens and intestines of lead chloride-exposed pigs were more severe than those of sodium chloride-exposed controls in experiment 3 is probably a manifestation of more severe illness in these pigs. The observation that intestinal lesions in lead chloride-exposed pigs were more severe than those of sodium chloride-exposed controls may indicate that bacteria were allowed to multiply more freely in the intestinal tract of the former. It must be remembered, however, that such observations are very crude measurements.

Microscopic lesions in experiments 2 and 3 were those expected in salmonellosis.

## SUMMARY

The effects of subclinical levels of lead exposure on the resistance of swine to Salmonella choleraesuis var. Kunzendorf were studied in 2 separate experiments (experiments 2 and 3). Before such studies could be performed, however, it was necessary to identify levels of lead exposure which would cause clinical and subclinical toxicosis in the pig. Experiment 1 was performed for this purpose.

### Experiment 1

Thirty pigs were divided into 10 groups of 3 pigs per group. Four groups received lead acetate orally at 4 different dosages, and 4 groups received lead acetate intraperitoneally (i.p.) at 4 different dosages. Two groups served as oral- and i.p.-control groups, and each pig in these groups received a different level of sodium acetate. Lead or sodium acetate was administered 6 days per week for 13 weeks (90 days).

It was demonstrated that pigs are extremely tolerant of lead. Blood lead concentrations were above 100 ug/dl in several pigs and reached 14,300 ug/dl in 1 pig, but these pigs did not die of lead toxicosis. Only mild clinical signs of toxicosis (gauntness and rough coats) were detected in orally lead acetate-exposed pigs. In pigs which received

lead acetate i.p., signs of lead toxicosis, if they occurred, were masked by signs resulting from lead acetate-induced peritonitis.

Significant decreases in blood hemoglobin concentrations, mean corpuscular hemoglobin concentrations (MCHCs), and/or mean corpuscular hemoglobins (MCHs) occurred in several i.p. lead acetate-exposed groups and were probably the result of a lead-induced reduction of blood ALAD activities. Basophilic stippling was observed in erythrocytes of a few lead acetate-exposed pigs but not in those of sodium acetate-exposed controls.

The only alterations of total leukocyte or leukocyte differential counts related to lead exposure occurred in response to peritonitis induced by i.p. injections of lead acetate solutions.

Decreases in serum albumin concentrations and increases in serum gamma globulin concentrations appeared to be the only serum protein alterations related to lead exposure. Serum gamma globulin concentrations did not decrease in any lead acetate-exposed groups. Decreased albumin concentrations may have resulted from subclinical lead-induced liver injury or from decreased feed intake. Increased gamma globulin concentrations probably occurred as a result of antigenic stimulation caused by i.p. injections of irritating lead acetate solutions.

As expected, high lead concentrations were detected in bone, liver, and kidney, but high concentrations also occurred in spleen, lung, and bone marrow of various groups. Lymph nodes of orally lead acetate-exposed pigs had low lead concentrations, and if lead were to significantly interfere with antibody synthesis at this site, it would have to do so at very low concentrations.

Peritonitis, which was centered at injection sites, was evident macroscopically in most i.p. lead acetate-exposed pigs.

Microscopic lesions associated with lead acetate-exposure were acid-fast intranuclear inclusion bodies found in livers and/or kidneys of a small number of orally- or i.p.-exposed pigs, and granulomatous peritonitis and lymphadenitis associated with lead acetate deposits in i.p.-exposed pigs.

## Experiments 2 and 3

In experiment 2, 36 pigs were divided into 2 groups of 18 pigs each. One group was fed a ration containing 500 ppm lead acetate while the second was fed a ration containing sodium acetate. After the first week of lead exposure and at weekly intervals thereafter, 6 pigs from each group were challenged i.p. with live S. choleraesuis var. Kunzendorf organisms.

In experiment 3, 24 pigs were divided into 3 groups of 8 pigs each. Two groups were fed a ration containing 3,000 ppm lead chloride for 1 and 2 weeks, respectively, and 1 group was fed a ration containing sodium chloride for 2 weeks. At the end of the lead or sodium chloride-exposure period, all pigs were challenged orally with live S. choleraesuis var. Kunzendorf organisms.

Lead chloride-exposed pigs had significantly higher mortality rates than did sodium chloride-exposed controls in experiment 3. Only 1 pig died during experiment 2. Clinical signs of lead-exposed pigs did not appear to be more severe than those of control pigs in either experiment.

In experiment 3, pigs exposed to lead chloride for 2 weeks had reduced appetites near the end of the lead-exposure period, and both lead chloride-exposed groups had significant decreases in blood hemoglobin concentrations, MCHCs, and MCHs as compared to the sodium-chloride-control group. These findings could have resulted from mild lead toxicosis, and it might be invalid to classify the pigs as subclinically intoxicated.

Alterations in leukocyte kinetics following Salmonella challenge in experiment 3 seem to indicate that lead chloride-exposed pigs were being more intensely affected by endotoxin. This may have resulted from greater production of endotoxin in these pigs due to lead-induced suppression of the immune

response to Salmonella or from a lead induced hypersensitivity to endotoxin.

Macroscopic lesions of lead chloride-exposed pigs were more severe than those of sodium chloride-controls in experiment 3. This was probably a manifestation of more severe illness in these pigs.

Lead-exposed pigs in both experiments had significantly lower serum alpha globulin concentrations, but the cause and significance of this alteration are not clear.

Despite the fact that lead exposure in experiments 2 and 3 caused a marked biological effect (decreased blood ALAD activity), there was no evidence that such exposure caused an alteration of humoral immunity as measured by serum agglutinin titers or serum beta and gamma globulin concentrations. It is, therefore, doubtful that suppression of humoral immunity caused the decreased resistance of lead chloride-exposed pigs in experiment 3.

In light of reports that lead acetate may impair phagocytosis and that protective immunity against Salmonella infection is probably cellular in nature, an alteration in cellular immunity might be considered as a cause of lead-induced reduction of resistance to Salmonella infection. The responsiveness of the cellular immune system was not studied in these experiments.

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# APPENDIX

Table 36. Experiment 1--Mean blood lead concentrations (ug/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 5 | 4 | 8 | 6 | 11 |
| 1 | 28 | 33 | 31 | 38 | 7 |
| 2 | 28 | 32 | 34 | 50 | 9 |
| 3 | 39 | 46 | 48 | 68 | 13 |
| 4 | 30 | 37 | 34 | 58 | 8 |
| 5 | 38 | 44 | 50 | 82 | 12 |
| 6 | 41 | 35 | 52 | 71 | 11 |
| 7 | 25 | 28 | 38 | 53 | 7 |
| 8 | 44 | 46 | 60 | 86 | 15 |
| 9 | 38 | 38 | 63 | 69 | 13 |
| 10 | 44 | 39 | 79 | 91 | 6 |
| 11 | 43 | 35 | 80 | 105 | 10 |
| 12 | 40 | 36 | 61 | 79 | 8 |
| 13 | 57 | 46 | 77 | 164 | 8 |

[a]Prior to lead or sodium acetate exposure.

Table 37. Experiment 1--Mean blood lead concentrations (ug/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 6 | 5 | 5 | 5 | 9 |
| 1 | 30 | 33 | 35 | 35 | 8 |
| 2 | 30 | 36 | 44 | 42 | 10 |
| 3 | 39 | 51 | 58 | 208 | 11 |
| 4 | 43 | 55 | 56 | 146 | 14 |
| 5 | 56 | 76 | 89 | 59 | 17 |
| 6 | 47 | 61 | 57 | 122 | 11 |
| 7 | 68 | 88 | 83 | 185 | 9 |
| 8 | 90 | 124 | 133 | 395 | 24 |
| 9 | 59 | 79 | 93 | 599 | 13 |
| 10 | 79 | 92 | 110 | 710 | 12 |
| 11 | 110 | 156 | 38 | 2,800 | 9 |
| 12 | 79 | 83 | 110 | 5,800 | 10 |
| 13 | 101 | 136 | 150 | 14,300 | 14 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 38. Experiment 1--Mean blood ALAD activities of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 219.4 | 176.1 | 218.9 | 185.9 | 197.7 |
| 1 | 67.5 | 45.0 | 52.8 | 37.5 | 209.2 |
| 2 | 31.6 | 19.4 | 20.4 | 13.5 | 175.4 |
| 3 | 20.4 | 13.2 | 12.4 | 7.6 | 161.5 |
| 4 | 19.1 | 13.9 | 11.5 | 10.2 | 156.8 |
| 5 | 12.1 | 8.9 | 9.8 | 7.7 | 109.6 |
| 6 | 15.0 | 13.2 | 11.7 | 9.0 | 153.4 |
| 7 | 7.8 | 6.5 | 5.4 | 4.7 | 107.1 |
| 8 | 5.3 | 4.4 | 3.3 | 3.3 | 123.6 |
| 9 | 4.8 | 4.1 | 2.8 | 2.8 | 111.1 |
| 10 | 4.5 | 3.3 | 3.0 | 3.2 | 85.3 |
| 11 | 2.7 | 1.8 | 1.2 | 1.1 | 88.9 |
| 12 | 3.5 | 3.2 | 5.2 | 2.6 | 75.4 |
| 13 | 4.1 | 2.9 | 4.3 | 3.3 | 76.9 |

[a]Prior to lead or sodium acetate exposure.

Table 39. Experiment 1--Mean blood ALAD activities of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 216.8 | 219.0 | 270.9 | 196.7 | 242.2 |
| 1 | 58.9 | 79.8 | 66.9 | 55.1 | 180.7 |
| 2 | 32.4 | 29.8 | 21.2 | 19.2 | 154.1 |
| 3 | 18.7 | 18.8 | 15.9 | 9.2 | 131.1 |
| 4 | 16.2 | 13.2 | 19.4 | 12.2 | 94.2 |
| 5 | 8.3 | 6.4 | 7.8 | 7.2 | 74.3 |
| 6 | 10.6 | 10.1 | 8.1 | 8.3 | 81.3 |
| 7 | 6.3 | 3.4 | 6.9 | 4.5 | 65.6 |
| 8 | 4.8 | 3.6 | 5.1 | 6.2 | 67.0 |
| 9 | 1.4 | 2.2 | 0.0 | 0.0 | 55.6 |
| 10 | 3.1 | 2.2 | 2.2 | 4.5 | 36.7 |
| 11 | 2.0 | 1.5 | 2.5 | 4.0 | 39.6 |
| 12 | 1.5 | 1.5 | 2.2 | 8.0 | 30.5 |
| 13 | 1.8 | 3.0 | 3.3 | 6.2 | 37.1 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 40. Experiment 1--Mean blood hemoglobin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 11.9 | 12.4 | 12.6 | 12.6 | 12.7 |
| 1 | 11.9 | 11.8 | 11.4 | 11.8 | 11.8 |
| 2 | 12.5 | 13.5 | 12.6 | 11.1 | 12.9 |
| 3 | 12.9 | 13.6 | 13.8 | 11.9 | 13.5 |
| 4 | 12.1 | 12.3 | 12.8 | 12.1 | 12.0 |
| 5 | 12.1 | 11.9 | 12.1 | 12.1 | 12.6 |
| 6 | 11.8 | 11.8 | 11.4 | 11.6 | 12.2 |
| 7 | 11.8 | 12.5 | 11.8 | 11.5 | 12.3 |
| 8 | 12.8 | 13.2 | 12.7 | 12.2 | 13.1 |
| 9 | 12.1 | 12.1 | 12.6 | 11.3 | 12.8 |
| 10 | 12.6 | 12.4 | 12.8 | 11.5 | 12.8 |
| 11 | 12.4 | 12.6 | 12.8 | 11.3 | 11.1 |
| 12 | 11.8 | 12.3 | 12.3 | 11.5 | 12.5 |
| 13 | 12.4 | 12.4 | 10.8 | 12.3 | 12.3 |

[a]Prior to lead or sodium acetate exposure.

Table 41. Experiment 1--Mean blood hemoglobin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 12.3 | 12.4 | 10.8 | 13.1 | 11.9 |
| 1 | 11.1 | 11.4 | 10.8 | 11.0 | 11.9 |
| 2 | 11.4 | 12.1 | 10.9 | 11.2 | 13.2 |
| 3 | 11.6 | 12.1 | 11.2 | 11.7 | 13.4 |
| 4 | 10.9 | 12.0 | 10.2 | 10.2 | 12.8 |
| 5 | 10.2 | 10.8 | 11.2 | 10.6 | 12.6 |
| 6 | 11.1 | 11.1 | 9.8 | 8.9 | 11.9 |
| 7 | 10.7 | 11.1 | 10.1 | 8.3 | 11.7 |
| 8 | 11.8 | 12.0 | 10.4 | 8.0 | 13.1 |
| 9 | 11.9 | 11.8 | 11.1 | 7.4 | 12.0 |
| 10 | 11.0 | 10.4 | 9.8 | 6.4 | 12.5 |
| 11 | 11.2 | 11.7 | 8.6 | 7.4 | 12.4 |
| 12 | 11.2 | 10.9 | 8.9 | 7.7 | 12.9 |
| 13 | 10.9 | 10.9 | 8.6 | 8.3 | 12.6 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 42. Experiment 1--Mean hematocrit percentages of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 38.3 | 39.3 | 39.8 | 40.3 | 39.5 |
| 1 | 36.7 | 37.7 | 35.7 | 37.5 | 38.7 |
| 2 | 39.8 | 42.7 | 41.0 | 37.0 | 41.0 |
| 3 | 41.0 | 41.0 | 41.0 | 38.7 | 41.0 |
| 4 | 37.0 | 36.7 | 39.7 | 36.3 | 37.0 |
| 5 | 37.7 | 37.7 | 38.0 | 38.0 | 40.0 |
| 6 | 36.3 | 38.3 | 35.7 | 37.3 | 37.3 |
| 7 | 37.3 | 38.7 | 38.0 | 35.7 | 37.7 |
| 8 | 41.0 | 39.0 | 39.0 | 38.3 | 41.3 |
| 9 | 35.7 | 36.7 | 37.7 | 34.7 | 37.0 |
| 10 | 38.7 | 39.0 | 39.0 | 36.3 | 41.0 |
| 11 | 37.7 | 39.3 | 39.3 | 35.3 | 33.0 |
| 12 | 34.7 | 37.0 | 36.3 | 35.7 | 36.7 |
| 13 | 40.0 | 40.3 | 35.7 | 40.7 | 40.0 |

[a]Prior to lead or sodium acetate exposure.

Table 43. Experiment 1--Mean hematocrit percentages of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 37.8 | 38.3 | 35.2 | 39.7 | 36.7 |
| 1 | 35.0 | 36.3 | 33.0 | 37.0 | 37.2 |
| 2 | 40.5 | 40.3 | 39.0 | 39.3 | 37.5 |
| 3 | 37.8 | 40.0 | 36.7 | 37.0 | 41.0 |
| 4 | 33.3 | 37.7 | 29.5 | 30.5 | 38.3 |
| 5 | 30.0 | 32.7 | 32.5 | 35.0 | 38.3 |
| 6 | 35.3 | 37.2 | 31.0 | 30.0 | 38.7 |
| 7 | 33.7 | 36.7 | 32.5 | 28.0 | 36.0 |
| 8 | 38.7 | 38.0 | 32.0 | 30.0 | 40.3 |
| 9 | 38.3 | 37.3 | 34.0 | 25.0 | 34.7 |
| 10 | 36.3 | 37.3 | 34.0 | 22.0 | 39.0 |
| 11 | 36.3 | 37.7 | 30.0 | 25.0 | 40.3 |
| 12 | 33.7 | 34.3 | 28.0 | 25.0 | 39.7 |
| 13 | 37.0 | 36.0 | 30.0 | 28.0 | 40.0 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 44. Experiment 1--Mean total red blood cell counts x $10^6$ (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 (control) |
| 0[a] | 7.08 | 7.12 | 7.41 | 7.54 | 8.58 |
| 1 | 6.74 | 7.14 | 7.11 | 7.45 | 7.04 |
| 2 | 8.43 | 8.83 | 7.30 | 7.45 | 7.14 |
| 3 | 7.19 | 7.75 | 7.25 | 7.79 | 6.43 |
| 4 | 7.01 | 7.99 | 6.80 | 7.06 | 8.75 |
| 5 | 5.71 | 5.26 | 5.31 | 6.04 | 6.94 |
| 6 | 6.29 | 6.95 | 5.88 | 6.80 | 6.50 |
| 7 | 7.04 | 6.80 | 6.85 | 6.52 | 6.43 |
| 8 | 6.84 | 6.17 | 6.79 | 6.54 | 7.35 |
| 9 | 7.37 | 7.22 | 7.26 | 6.81 | 7.61 |
| 10 | 6.84 | 7.30 | 7.06 | 7.15 | 7.92 |
| 11 | 5.48 | 5.89 | 5.86 | 5.34 | 4.80 |
| 12 | 7.49 | 7.40 | 7.42 | 7.66 | 7.80 |
| 13 | 7.21 | 6.78 | 6.21 | 7.83 | 6.48 |

[a]Prior to lead or sodium acetate exposure.

Table 45. Experiment 1--Mean total red blood cell counts x $10^6$ (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 7.87 | 7.88 | 7.28 | 8.79 | 7.65 |
| 1 | 7.04 | 7.86 | 6.76 | 6.37 | 6.70 |
| 2 | 6.70 | 7.29 | 6.71 | 6.59 | 7.68 |
| 3 | 5.98 | 6.75 | 5.99 | 6.20 | 6.10 |
| 4 | 5.54 | 6.23 | 6.22 | 5.38 | 6.43 |
| 5 | 6.19 | 6.96 | 6.84 | 7.48 | 6.42 |
| 6 | 6.06 | 6.17 | 5.31 | 5.27 | 6.03 |
| 7 | 6.75 | 6.55 | 6.26 | 5.23 | 6.61 |
| 8 | 6.84 | 6.35 | 6.41 | 4.84 | 6.84 |
| 9 | 7.36 | 6.40 | 7.27 | 5.19 | 6.63 |
| 10 | 6.80 | 6.48 | 6.99 | 4.25 | 7.18 |
| 11 | 5.57 | 5.86 | 5.56 | 4.30 | 5.92 |
| 12 | 7.45 | 7.04 | 5.93 | 5.32 | 7.28 |
| 13 | 6.10 | 5.74 | 5.18 | 4.82 | 6.72 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 46. Experiment 1--Mean MCVs (fl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 54.3 | 55.4 | 53.8 | 53.5 | 47.1 |
| 1 | 54.7 | 53.0 | 50.4 | 50.5 | 55.2 |
| 2 | 47.5 | 50.8 | 56.2 | 50.1 | 60.3 |
| 3 | 57.2 | 52.9 | 56.5 | 50.4 | 68.1 |
| 4 | 56.2 | 46.1 | 58.5 | 51.6 | 42.6 |
| 5 | 66.0 | 72.1 | 72.0 | 63.2 | 60.0 |
| 6 | 57.7 | 55.2 | 62.0 | 54.9 | 57.7 |
| 7 | 53.2 | 57.2 | 55.7 | 54.8 | 60.1 |
| 8 | 60.0 | 64.8 | 58.8 | 59.2 | 56.2 |
| 9 | 49.0 | 50.9 | 52.5 | 51.0 | 48.5 |
| 10 | 56.8 | 53.5 | 55.3 | 50.9 | 52.1 |
| 11 | 68.7 | 66.5 | 67.2 | 67.0 | 68.8 |
| 12 | 46.4 | 50.0 | 49.0 | 46.4 | 47.2 |
| 13 | 55.5 | 59.7 | 58.3 | 52.9 | 61.7 |

[a]Prior to lead or sodium acetate exposure.

Table 47. Experiment 1--Mean MCVs (fl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 49.4 | 49.0 | 49.7 | 45.9 | 47.8 |
| 1 | 50.0 | 46.6 | 49.2 | 58.1 | 55.9 |
| 2 | 60.5 | 55.4 | 58.2 | 59.7 | 48.8 |
| 3 | 63.4 | 60.7 | 61.3 | 60.0 | 68.3 |
| 4 | 62.9 | 60.7 | 47.5 | 56.8 | 60.4 |
| 5 | 48.8 | 47.0 | 48.5 | 47.1 | 60.0 |
| 6 | 58.5 | 60.4 | 58.4 | 56.9 | 64.2 |
| 7 | 51.1 | 56.3 | 52.3 | 53.5 | 54.5 |
| 8 | 56.7 | 59.8 | 49.9 | 62.0 | 59.0 |
| 9 | 52.4 | 58.6 | 46.8 | 48.2 | 52.3 |
| 10 | 53.5 | 57.9 | 48.6 | 51.8 | 55.0 |
| 11 | 66.3 | 64.6 | 54.0 | 58.1 | 68.1 |
| 12 | 45.7 | 48.9 | 47.2 | 47.0 | 54.4 |
| 13 | 60.8 | 63.2 | 57.9 | 58.1 | 59.8 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 48. Experiment 1--Mean MCHCs (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 31.0 | 31.4 | 31.6 | 31.2 | 32.2 |
| 1 | 32.4 | 31.3 | 32.1 | 31.4 | 30.4 |
| 2 | 31.3 | 31.6 | 30.8 | 30.1 | 31.4 |
| 3 | 31.5 | 33.2 | 33.7 | 30.7 | 31.1 |
| 4 | 32.7 | 33.5 | 32.4 | 33.2 | 32.4 |
| 5 | 32.0 | 31.6 | 31.9 | 31.9 | 32.0 |
| 6 | 32.5 | 30.8 | 32.0 | 31.2 | 32.8 |
| 7 | 31.5 | 32.4 | 31.0 | 32.4 | 32.7 |
| 8 | 31.1 | 33.9 | 32.6 | 31.7 | 31.8 |
| 9 | 34.0 | 33.2 | 33.4 | 32.6 | 35.0 |
| 10 | 32.6 | 31.8 | 32.9 | 31.8 | 31.3 |
| 11 | 32.8 | 31.9 | 32.6 | 32.1 | 33.7 |
| 12 | 34.0 | 33.2 | 33.8 | 32.6 | 34.0 |
| 13 | 31.1 | 30.7 | 30.1 | 30.2 | 30.7 |

[a]Prior to lead or sodium acetate exposure.

Table 49. Experiment 1--Mean MCHCs (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 32.4 | 32.3 | 30.8 | 32.9 | 32.5 |
| 1 | 31.6 | 31.5 | 32.6 | 29.7 | 32.0 |
| 2 | 28.2 | 30.1 | 28.1 | 28.5 | 35.2 |
| 3 | 30.7 | 30.2 | 30.5 | 31.5 | 32.6 |
| 4 | 32.8 | 31.8 | 34.7 | 33.6 | 33.3 |
| 5 | 34.0 | 32.8 | 34.8 | 30.3 | 32.9 |
| 6 | 31.4 | 29.8 | 31.6 | 29.7 | 30.8 |
| 7 | 32.1 | 30.2 | 31.2 | 29.6 | 32.4 |
| 8 | 30.6 | 31.5 | 32.5 | 26.7 | 32.4 |
| 9 | 31.0 | 31.5 | 32.6 | 29.6 | 34.9 |
| 10 | 30.3 | 28.0 | 28.8 | 29.1 | 32.2 |
| 11 | 30.8 | 31.0 | 28.7 | 29.6 | 30.7 |
| 12 | 33.2 | 31.6 | 31.8 | 30.8 | 32.4 |
| 13 | 29.5 | 30.2 | 28.7 | 29.6 | 31.4 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 50. Experiment 1--Mean MCHs (pg) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 16.8 | 17.4 | 17.0 | 16.7 | 15.1 |
| 1 | 17.8 | 16.6 | 16.1 | 15.8 | 16.8 |
| 2 | 14.8 | 16.0 | 17.3 | 15.1 | 18.7 |
| 3 | 18.0 | 17.6 | 19.0 | 15.5 | 21.2 |
| 4 | 18.3 | 15.4 | 19.0 | 17.2 | 13.8 |
| 5 | 21.1 | 22.8 | 22.9 | 20.2 | 18.2 |
| 6 | 18.8 | 17.0 | 19.8 | 17.1 | 18.9 |
| 7 | 16.8 | 18.5 | 17.4 | 17.7 | 19.4 |
| 8 | 18.7 | 21.8 | 19.2 | 18.8 | 17.9 |
| 9 | 16.7 | 16.9 | 17.6 | 16.6 | 16.9 |
| 10 | 18.5 | 17.0 | 18.2 | 16.2 | 16.3 |
| 11 | 22.6 | 21.2 | 21.9 | 21.6 | 23.2 |
| 12 | 15.8 | 16.6 | 16.5 | 15.1 | 16.1 |
| 13 | 17.3 | 18.3 | 17.6 | 16.0 | 18.9 |

[a]Prior to lead or sodium acetate exposure.

Table 51. Experiment 1--Mean MCHs (pg) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 15.9 | 15.8 | 15.3 | 15.1 | 15.6 |
| 1 | 15.8 | 14.7 | 16.0 | 17.2 | 17.4 |
| 2 | 17.0 | 16.7 | 16.3 | 17.0 | 17.2 |
| 3 | 19.5 | 18.3 | 18.6 | 18.9 | 22.3 |
| 4 | 20.7 | 19.3 | 16.5 | 19.1 | 20.2 |
| 5 | 16.6 | 15.4 | 16.6 | 14.2 | 19.8 |
| 6 | 18.3 | 18.0 | 18.4 | 16.9 | 19.8 |
| 7 | 16.2 | 17.0 | 16.2 | 15.9 | 17.7 |
| 8 | 17.4 | 18.8 | 16.2 | 16.5 | 19.1 |
| 9 | 16.2 | 18.5 | 15.3 | 14.2 | 18.2 |
| 10 | 16.2 | 16.2 | 14.0 | 15.0 | 17.6 |
| 11 | 20.4 | 20.0 | 15.5 | 17.2 | 20.9 |
| 12 | 15.2 | 15.5 | 15.0 | 14.5 | 17.6 |
| 13 | 18.0 | 19.1 | 16.6 | 17.2 | 18.8 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 52. Experiment 1--Mean total leukocyte counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 27,433 | 26,433 | 23,733 | 23,300 | 22,866 |
| 1 | 22,600 | 21,067 | 19,600 | 24,567 | 22,567 |
| 2 | 21,033 | 21,600 | 20,767 | 22,467 | 19,533 |
| 3 | 21,800 | 17,433 | 22,133 | 22,300 | 15,267 |
| 4 | 20,833 | 20,367 | 19,733 | 19,833 | 22,500 |
| 5 | 20,300 | 18,367 | 16,700 | 20,800 | 21,967 |
| 6 | 22,533 | 18,833 | 17,133 | 24,933 | 21,933 |
| 7 | 22,167 | 18,767 | 20,333 | 20,333 | 16,767 |
| 8 | 20,133 | 20,467 | 19,133 | 20,533 | 18,267 |
| 9 | 19,933 | 21,700 | 20,667 | 23,300 | 17,833 |
| 10 | 17,933 | 13,367 | 17,633 | 21,467 | 17,133 |
| 11 | 12,333 | 10,333 | 11,900 | 11,167 | 11,533 |
| 12 | 17,033 | 16,400 | 17,000 | 19,200 | 17,067 |
| 13 | 17,267 | 14,400 | 14,367 | 19,000 | 15,733 |

[a]Prior to lead or sodium acetate exposure.

Table 53. Experiment 1--Mean total leukocyte counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group 6 | 7 | 8[a] | 9[a] | 10 (control) |
|---|---|---|---|---|---|
| 0[b] | 23,067 | 26,667 | 24,933 | 21,500 | 18,167 |
| 1 | 20,967 | 22,767 | 22,133 | 19,667 | 24,700 |
| 2 | 23,650 | 19,433 | 22,267 | 24,633 | 21,967 |
| 3 | 16,367 | 25,367 | 19,267 | 22,933 | 16,867 |
| 4 | 17,300 | 22,767 | 20,000 | 28,300 | 20,567 |
| 5 | 21,800 | 20,500 | 23,350 | 24,400 | 22,367 |
| 6 | 19,833 | 19,667 | 16,900 | 26,200 | 20,533 |
| 7 | 16,067 | 21,967 | 18,500 | 26,300 | 21,033 |
| 8 | 20,033 | 20,833 | 16,700 | 26,600 | 21,367 |
| 9 | 21,433 | 19,967 | 16,800 | 20,600 | 20,833 |
| 10 | 20,533 | 20,333 | 18,400 | 43,500 | 18,433 |
| 11 | 15,933 | 19,967 | 15,200 | 28,700 | 16,400 |
| 12 | 18,033 | 18,967 | 14,200 | 27,700 | 19,433 |
| 13 | 16,467 | 20,067 | 16,800 | 15,000 | 18,500 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 54. Experiment 1--Mean total segmented neutrophil counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 5,331 | 10,119 | 7,961 | 8,737 | 8,175 |
| 1 | 7,221 | 7,536 | 5,244 | 8,121 | 8,375 |
| 2 | 5,250 | 8,003 | 5,128 | 7,844 | 7,104 |
| 3 | 5,049 | 4,804 | 4,956 | 6,645 | 4,989 |
| 4 | 5,923 | 7,835 | 5,401 | 5,839 | 11,074 |
| 5 | 6,902 | 7,620 | 3,575 | 6,558 | 9,845 |
| 6 | 4,956 | 5,528 | 4,273 | 8,122 | 9,020 |
| 7 | 5,347 | 5,762 | 5,901 | 5,328 | 4,867 |
| 8 | 6,482 | 6,845 | 4,034 | 3,632 | 5,934 |
| 9 | 4,281 | 5,639 | 4,236 | 7,108 | 5,327 |
| 10 | 5,537 | 4,552 | 4,831 | 6,743 | 6,055 |
| 11 | 2,495 | 2,767 | 2,851 | 1,904 | 4,153 |
| 12 | 4,987 | 3,737 | 4,560 | 5,517 | 5,028 |
| 13 | 3,215 | 2,603 | 2,167 | 3,627 | 3,352 |

[a]Prior to lead or sodium acetate exposure.

Table 55. Experiment 1--Mean total segmented neutrophil counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 6,090 | 10,503 | 10,067 | 6,640 | 7,728 |
| 1 | 6,250 | 9,155 | 8,029 | 5,419 | 9,182 |
| 2 | 8,128 | 7,585 | 7,313 | 9,240 | 8,052 |
| 3 | 4,828 | 12,274 | 6,893 | 6,581 | 3,902 |
| 4 | 5,571 | 7,673 | 7,616 | 12,768 | 6,786 |
| 5 | 8,213 | 8,083 | 9,020 | 7,086 | 6,706 |
| 6 | 4,309 | 4,771 | 7,605 | 7,086 | 5,393 |
| 7 | 3,628 | 6,756 | 6,529 | 7,627 | 5,923 |
| 8 | 5,341 | 8,441 | 6,513 | 14,896 | 4,522 |
| 9 | 6,865 | 5,703 | 3,192 | 7,622 | 4,560 |
| 10 | 5,417 | 5,823 | 6,808 | 22,185 | 5,190 |
| 11 | 3,828 | 5,215 | 4,712 | 12,341 | 3,883 |
| 12 | 3,737 | 5,227 | 4,260 | 11,357 | 4,991 |
| 13 | 2,615 | 4,458 | 6,384 | 8,250 | 4,152 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 56. Experiment 1--Mean total band neutrophil counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | (control) |
|---|---|---|---|---|---|
| 0[a] | 620 | 2,687 | 483 | 881 | 1,733 |
| 1 | 1,053 | 2,485 | 865 | 1,829 | 2,107 |
| 2 | 354 | 776 | 209 | 674 | 751 |
| 3 | 940 | 1,806 | 1,074 | 385 | 655 |
| 4 | 1,028 | 2,675 | 884 | 559 | 1,350 |
| 5 | 963 | 1,885 | 723 | 1,059 | 1,368 |
| 6 | 594 | 1,105 | 674 | 1,428 | 1,187 |
| 7 | 581 | 2,052 | 1,112 | 1,052 | 734 |
| 8 | 638 | 3,574 | 751 | 1,243 | 445 |
| 9 | 1,194 | 3,552 | 911 | 694 | 780 |
| 10 | 163 | 440 | 477 | 239 | 106 |
| 11 | 263 | 802 | 374 | 378 | 500 |
| 12 | 576 | 1,711 | 351 | 797 | 1,337 |
| 13 | 658 | 1,919 | 1,324 | 891 | 791 |

[a]Prior to lead or sodium acetate exposure.

Table 57. Experiment 1--Mean total band neutrophil counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 1,236 | 1,268 | 1,259 | 1,918 | 625 |
| 1 | 2,830 | 2,288 | 1,847 | 3,661 | 2,169 |
| 2 | 818 | 595 | 1,594 | 1,701 | 355 |
| 3 | 925 | 2,580 | 912 | 4,251 | 1,245 |
| 4 | 1,495 | 905 | 2,608 | 3,462 | 2,330 |
| 5 | 719 | 2,920 | 2,292 | 9,312 | 2,999 |
| 6 | 1,320 | 1,762 | 1,521 | 8,646 | 2,080 |
| 7 | 1,298 | 1,960 | 1,672 | 7,101 | 2,028 |
| 8 | 654 | 1,488 | 1,670 | 1,596 | 1,374 |
| 9 | 1,024 | 1,264 | 2,352 | 5,974 | 2,401 |
| 10 | 689 | 941 | 1,656 | 6,090 | 3,817 |
| 11 | 450 | 974 | 912 | 3,731 | 908 |
| 12 | 881 | 989 | 852 | 5,817 | 1,436 |
| 13 | 642 | 1,540 | 1,848 | 2,250 | 1,516 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 58. Experiment 1--Mean total lymphocyte counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 19,005 | 12,277 | 14,015 | 12,833 | 11,783 |
| 1 | 12,862 | 9,376 | 12,119 | 12,274 | 10,773 |
| 2 | 14,799 | 11,736 | 13,916 | 12,416 | 10,702 |
| 3 | 15,183 | 9,668 | 15,025 | 14,568 | 8,594 |
| 4 | 12,306 | 8,394 | 12,190 | 11,291 | 8,790 |
| 5 | 11,459 | 8,272 | 11,058 | 11,785 | 9,586 |
| 6 | 15,949 | 10,892 | 11,082 | 14,007 | 11,207 |
| 7 | 15,669 | 9,785 | 12,320 | 12,662 | 10,076 |
| 8 | 11,890 | 7,910 | 12,441 | 14,445 | 11,045 |
| 9 | 13,667 | 9,020 | 13,872 | 14,058 | 11,037 |
| 10 | 11,515 | 7,718 | 11,495 | 12,749 | 10,532 |
| 11 | 9,139 | 6,510 | 8,083 | 8,264 | 6,659 |
| 12 | 10,823 | 9,876 | 11,365 | 12,090 | 10,056 |
| 13 | 12,684 | 9,056 | 10,325 | 13,277 | 10,983 |

[a]Prior to lead or sodium acetate exposure.

Table 59. Experiment 1--Mean total lymphocyte counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 9,759 | 12,768 | 12,743 | 11,738 | 8,656 |
| 1 | 9,722 | 9,455 | 10,437 | 9,186 | 12,366 |
| 2 | 12,581 | 10,407 | 12,194 | 12,393 | 12,373 |
| 3 | 10,173 | 9,275 | 10,497 | 11,255 | 10,381 |
| 4 | 9,771 | 12,770 | 8,992 | 10,870 | 9,679 |
| 5 | 11,260 | 8,811 | 10,839 | 7,628 | 11,840 |
| 6 | 13,000 | 11,103 | 7,098 | 9,432 | 11,862 |
| 7 | 10,326 | 10,790 | 9,205 | 11,572 | 11,702 |
| 8 | 11,950 | 8,526 | 7,181 | 9,310 | 13,954 |
| 9 | 12,379 | 10,704 | 10,584 | 6,798 | 12,823 |
| 10 | 12,717 | 11,734 | 8,648 | 14,355 | 11,773 |
| 11 | 11,044 | 12,650 | 8,360 | 11,480 | 11,033 |
| 12 | 12,337 | 11,243 | 8,236 | 10,526 | 12,424 |
| 13 | 11,986 | 11,912 | 7,392 | 3,750 | 11,679 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 60. Experiment 1--Mean total eosinophil counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 780 | 769 | 186 | 373 | 287 |
| 1 | 295 | 486 | 838 | 321 | 485 |
| 2 | 417 | 306 | 944 | 648 | 411 |
| 3 | 134 | 427 | 672 | 280 | 268 |
| 4 | 678 | 871 | 570 | 797 | 363 |
| 5 | 209 | 290 | 727 | 503 | 340 |
| 6 | 768 | 1,178 | 795 | 1,031 | 327 |
| 7 | 430 | 641 | 322 | 676 | 838 |
| 8 | 691 | 1,271 | 1,438 | 879 | 433 |
| 9 | 507 | 920 | 816 | 1,062 | 538 |
| 10 | 501 | 347 | 388 | 1,128 | 199 |
| 11 | 312 | 213 | 383 | 452 | 29 |
| 12 | 429 | 517 | 374 | 604 | 423 |
| 13 | 541 | 538 | 312 | 1,022 | 415 |

[a]Prior to lead or sodium acetate exposure.

Table 61. Experiment 1--Mean total eosinophil counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group 6 | 7 | 8[a] | 9[a] | 10 (control) |
|---|---|---|---|---|---|
| 0[b] | 162 | 775 | 311 | 225 | 224 |
| 1 | 709 | 583 | 414 | 536 | 661 |
| 2 | 879 | 293 | 378 | 350 | 599 |
| 3 | 167 | 152 | 116 | 65 | 397 |
| 4 | 231 | 807 | 200 | 400 | 206 |
| 5 | 932 | 304 | 129 | 260 | 379 |
| 6 | 729 | 1,158 | 676 | 0 | 869 |
| 7 | 715 | 1,953 | 440 | 0 | 825 |
| 8 | 1,235 | 1,524 | 167 | 0 | 915 |
| 9 | 791 | 1,788 | 336 | 0 | 782 |
| 10 | 1,124 | 1,529 | 736 | 870 | 567 |
| 11 | 568 | 921 | 912 | 861 | 260 |
| 12 | 956 | 1,069 | 710 | 0 | 450 |
| 13 | 390 | 1,682 | 504 | 450 | 1,096 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 62. Experiment 1--Mean total monocyte counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 1,425 | 582 | 710 | 373 | 810 |
| 1 | 1,013 | 929 | 359 | 1,700 | 771 |
| 2 | 142 | 640 | 514 | 752 | 566 |
| 3 | 377 | 664 | 354 | 421 | 639 |
| 4 | 765 | 536 | 613 | 1,148 | 859 |
| 5 | 557 | 300 | 450 | 643 | 736 |
| 6 | 267 | 130 | 271 | 345 | 193 |
| 7 | 70 | 527 | 379 | 553 | 193 |
| 8 | 274 | 666 | 470 | 335 | 399 |
| 9 | 285 | 217 | 620 | 377 | 102 |
| 10 | 109 | 223 | 318 | 441 | 65 |
| 11 | 38 | 41 | 165 | 125 | 192 |
| 12 | 153 | 509 | 295 | 61 | 169 |
| 13 | 112 | 237 | 119 | 124 | 192 |

[a]Prior to lead or sodium acetate exposure.

Table 63. Experiment 1--Mean total monocyte counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 1,273 | 955 | 554 | 979 | 644 |
| 1 | 1,306 | 1,086 | 1,184 | 800 | 239 |
| 2 | 1,008 | 434 | 723 | 864 | 524 |
| 3 | 227 | 905 | 741 | 653 | 712 |
| 4 | 164 | 612 | 488 | 799 | 1,567 |
| 5 | 393 | 268 | 1,070 | 114 | 365 |
| 6 | 474 | 798 | 0 | 262 | 329 |
| 7 | 100 | 437 | 0 | 0 | 404 |
| 8 | 601 | 712 | 835 | 798 | 602 |
| 9 | 295 | 432 | 336 | 206 | 59 |
| 10 | 234 | 184 | 552 | 0 | 324 |
| 11 | 43 | 0 | 304 | 0 | 118 |
| 12 | 68 | 121 | 142 | 0 | 63 |
| 13 | 165 | 270 | 672 | 300 | 56 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 64. Experiment 1--Mean total basophil counts (per cubic mm) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 272 | 0 | 186 | 102 | 289 |
| 1 | 157 | 255 | 175 | 321 | 55 |
| 2 | 70 | 139 | 55 | 133 | 0 |
| 3 | 117 | 65 | 53 | 0 | 122 |
| 4 | 134 | 56 | 0 | 56 | 64 |
| 5 | 209 | 0 | 167 | 151 | 92 |
| 6 | 0 | 0 | 38 | 0 | 0 |
| 7 | 70 | 0 | 0 | 62 | 59 |
| 8 | 159 | 200 | 0 | 0 | 0 |
| 9 | 0 | 151 | 212 | 0 | 50 |
| 10 | 108 | 87 | 126 | 167 | 175 |
| 11 | 85 | 0 | 45 | 43 | 0 |
| 12 | 65 | 49 | 54 | 131 | 53 |
| 13 | 56 | 47 | 119 | 59 | 0 |

[a]Prior to lead or sodium acetate exposure.

Table 65. Experiment 1--Mean total basophil counts (per cubic mm) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 0 | 297 | 0 | 0 | 41 |
| 1 | 149 | 199 | 222 | 0 | 83 |
| 2 | 0 | 120 | 65 | 85 | 142 |
| 3 | 46 | 181 | 107 | 129 | 229 |
| 4 | 69 | 0 | 96 | 0 | 0 |
| 5 | 283 | 114 | 0 | 0 | 78 |
| 6 | 0 | 74 | 0 | 0 | 0 |
| 7 | 0 | 0 | 0 | 0 | 151 |
| 8 | 252 | 143 | 334 | 0 | 0 |
| 9 | 79 | 76 | 0 | 0 | 208 |
| 10 | 352 | 122 | 0 | 0 | 198 |
| 11 | 0 | 207 | 0 | 287 | 199 |
| 12 | 54 | 318 | 0 | 0 | 69 |
| 13 | 598 | 204 | 0 | 0 | 0 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 66. Experiment 1--Mean plasma protein:fibrinogen ratios of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 20 | 22 | 21 | 34 | 20 |
| 1 | 15 | 20 | 18 | 22 | 19 |
| 2 | 25 | 21 | 28 | 33 | 20 |
| 3 | 30 | 25 | 24 | 44 | 16 |
| 4 | 43 | 41 | 34 | 40 | 16 |
| 5 | 42 | 29 | 31 | 29 | 22 |
| 6 | 29 | 25 | 23 | 40 | 28 |
| 7 | 28 | 31 | 27 | 27 | 29 |
| 8 | 19 | 29 | 27 | 25 | 24 |
| 9 | 39 | 30 | 26 | 39 | 44 |
| 10 | 33 | 60 | 37 | 39 | 31 |
| 11 | 64 | 59 | 60 | 63 | 56 |
| 12 | 52 | 72 | 40 | 37 | 66 |
| 13 | 20 | 27 | 22 | 29 | 35 |

[a]Prior to lead or sodium acetate exposure.

Table 67. Experiment 1--Mean plasma protein: fibrinogen ratios of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 20 | 20 | 21 | 32 | 29 |
| 1 | 14 | 18 | 21 | 16 | 17 |
| 2 | 18 | 13 | 31 | 31 | 64 |
| 3 | 31 | 25 | 40 | 30 | 39 |
| 4 | 19 | 21 | 35 | 19 | 19 |
| 5 | 17 | 23 | 15 | 23 | 28 |
| 6 | 35 | 26 | 66 | 10 | 26 |
| 7 | 37 | 31 | 42 | 21 | 24 |
| 8 | 18 | 45 | 34 | 20 | 38 |
| 9 | 39 | 56 | 38 | 32 | 28 |
| 10 | 30 | 45 | 27 | 31 | 58 |
| 11 | 40 | 70 | 71 | 34 | 29 |
| 12 | 54 | 54 | 65 | 22 | 72 |
| 13 | 39 | 56 | 72 | 18 | 20 |

[a]Values for weeks 4 and 5 are means from 2 pigs, and values for weeks 6-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 68. Experiment 1--Mean total serum protein concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 6.0 | 5.9 | 6.0 | 6.3 | 6.3 |
| 1 | 6.1 | 6.0 | 5.5 | 5.8 | 5.9 |
| 2 | 6.3 | 6.5 | 6.1 | 6.3 | 6.2 |
| 3 | 6.6 | 6.7 | 6.4 | 6.4 | 6.6 |
| 5 | 6.2 | 6.7 | 5.9 | 6.4 | 6.3 |
| 7 | 6.1 | 6.8 | 5.9 | 6.0 | 6.2 |
| 8 | 7.1 | 6.2 | 5.9 | 6.2 | 6.4 |
| 9 | 6.1 | 6.6 | 6.1 | 6.3 | 6.5 |
| 10 | 6.4 | 7.1 | 5.9 | 6.2 | 6.9 |
| 11 | 6.3 | 7.0 | 5.9 | 6.2 | 6.6 |
| 12 | 6.1 | 7.1 | 5.9 | 6.1 | 6.5 |
| 13 | 6.2 | 6.7 | 5.8 | 6.4 | 6.5 |

[a]Prior to lead or sodium acetate exposure.

Table 69. Experiment 1--Mean total serum protein concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 6.5 | 6.3 | 6.2 | 6.3 | 6.3 |
| 1 | 6.1 | 6.3 | 5.8 | 6.3 | 6.1 |
| 2 | 6.8 | 6.6 | 6.5 | 6.0 | 6.3 |
| 3 | 6.4 | 6.6 | 6.6 | 6.3 | 6.7 |
| 5 | 6.0 | 6.3 | 6.4 | 6.2 | 6.6 |
| 7 | 6.2 | 6.3 | --- | 5.9 | 6.1 |
| 8 | 6.2 | 6.6 | 6.7 | 5.7 | 6.8 |
| 9 | 6.3 | 6.6 | 7.3 | 6.1 | 6.3 |
| 10 | 6.4 | 6.7 | 7.7 | 6.0 | 6.7 |
| 11 | 6.5 | 6.9 | 7.0 | 6.6 | 7.1 |
| 12 | 6.3 | 6.5 | 6.4 | 6.3 | 7.1 |
| 13 | 6.2 | 6.6 | 7.1 | 7.0 | 6.4 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 70. Experiment 1--Mean serum albumin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 2.31 | 2.15 | 2.38 | 2.36 | 2.47 |
| 1 | 2.56 | 2.66 | 2.36 | 2.35 | 2.29 |
| 2 | 2.67 | 2.66 | 2.40 | 2.16 | 2.49 |
| 3 | 2.53 | 2.73 | 2.43 | 2.11 | 2.54 |
| 5 | 2.52 | 2.63 | 2.32 | 2.29 | 2.68 |
| 7 | 2.53 | 3.20 | 2.44 | 2.28 | 2.78 |
| 8 | 2.90 | 3.06 | 2.38 | 2.56 | 2.58 |
| 9 | 2.51 | 2.81 | 2.32 | 2.20 | 2.74 |
| 10 | 2.81 | 3.36 | 2.51 | 2.47 | 3.00 |
| 11 | 2.85 | 3.15 | 2.08 | 2.12 | 2.90 |
| 12 | 2.70 | 2.78 | 2.36 | 2.37 | 2.79 |
| 13 | 2.60 | 3.01 | 2.24 | 2.35 | 2.69 |

[a]Prior to lead or sodium acetate exposure.

Table 71. Experiment 1--Mean serum albumin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 2.56 | 2.26 | 2.09 | 2.38 | 2.41 |
| 1 | 2.21 | 2.29 | 1.97 | 2.22 | 2.35 |
| 2 | 2.64 | 2.19 | 2.36 | 1.98 | 2.51 |
| 3 | 2.52 | 2.27 | 2.12 | 1.95 | 2.62 |
| 5 | 2.21 | 1.93 | 1.85 | 1.90 | 2.56 |
| 7 | 2.33 | 2.00 | ---- | 1.66 | 2.37 |
| 8 | 2.26 | 1.97 | 2.26 | 1.65 | 3.15 |
| 9 | 2.35 | 2.10 | 2.11 | 1.77 | 2.46 |
| 10 | 3.06 | 2.53 | 2.62 | 1.76 | 2.97 |
| 11 | 2.56 | 2.29 | 1.95 | 1.70 | 2.90 |
| 12 | 2.12 | 2.42 | 1.96 | 1.58 | 2.81 |
| 13 | 2.42 | 2.38 | 2.31 | 2.12 | 2.46 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 72. Experiment 1--Mean serum alpha$_1$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 0.30 | 0.29 | 0.22 | 0.28 | 0.26 |
| 1 | 0.20 | 0.18 | 0.13 | 0.17 | 0.38 |
| 2 | 0.18 | 0.26 | 0.28 | 0.25 | 0.28 |
| 3 | 0.32 | 0.35 | 0.29 | 0.25 | 0.31 |
| 5 | 0.23 | 0.19 | 0.18 | 0.22 | 0.14 |
| 7 | 0.33 | 0.19 | 0.17 | 0.32 | 0.23 |
| 8 | 0.46 | 0.18 | 0.15 | 0.12 | 0.28 |
| 9 | 0.24 | 0.20 | 0.31 | 0.24 | 0.21 |
| 10 | 0.30 | 0.22 | 0.24 | 0.23 | 0.28 |
| 11 | 0.18 | 0.27 | 0.26 | 0.29 | 0.22 |
| 12 | 0.19 | 0.27 | 0.22 | 0.24 | 0.16 |
| 13 | 0.26 | 0.22 | 0.09 | 0.26 | 0.30 |

[a]Prior to lead or sodium acetate exposure.

Table 73. Experiment 1--Mean serum alpha$_1$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 0.28 | 0.32 | 0.32 | 0.26 | 0.29 |
| 1 | 0.21 | 0.27 | 0.29 | 0.26 | 0.21 |
| 2 | 0.18 | 0.24 | 0.29 | 0.24 | 0.22 |
| 3 | 0.22 | 0.25 | 0.27 | 0.28 | 0.20 |
| 5 | 0.19 | 0.18 | 0.33 | 0.20 | 0.18 |
| 7 | 0.24 | 0.26 | ---- | 0.26 | 0.39 |
| 8 | 0.26 | 0.30 | 0.22 | 0.20 | 0.17 |
| 9 | 0.28 | 0.27 | 0.33 | 0.34 | 0.19 |
| 10 | 0.18 | 0.24 | 0.15 | 0.12 | 0.28 |
| 11 | 0.23 | 0.33 | 0.19 | 0.13 | 0.33 |
| 12 | 0.25 | 0.19 | 0.36 | 0.13 | 0.24 |
| 13 | 0.18 | 0.21 | 0.29 | 0.31 | 0.24 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 74. Experiment 1--Mean serum alpha$_2$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 1.11 | 1.21 | 1.16 | 1.35 | 1.25 |
| 1 | 1.08 | 1.10 | 0.91 | 1.16 | 0.96 |
| 2 | 1.01 | 1.10 | 0.97 | 1.24 | 1.02 |
| 3 | 1.08 | 1.14 | 1.04 | 1.09 | 1.22 |
| 5 | 0.91 | 1.24 | 0.82 | 1.06 | 1.07 |
| 7 | 0.82 | 1.02 | 0.82 | 0.91 | 0.95 |
| 8 | 0.88 | 0.83 | 0.66 | 0.95 | 0.97 |
| 9 | 0.94 | 1.06 | 0.84 | 1.04 | 1.00 |
| 10 | 0.94 | 1.08 | 0.77 | 0.90 | 1.01 |
| 11 | 1.04 | 1.08 | 0.89 | 0.98 | 1.10 |
| 12 | 0.99 | 1.28 | 0.83 | 0.92 | 1.14 |
| 13 | 0.94 | 1.00 | 0.97 | 1.03 | 0.97 |

[a]Prior to lead or sodium acetate exposure.

Table 75. Experiment 1--Mean serum alpha$_2$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 1.19 | 1.16 | 1.24 | 1.30 | 1.14 |
| 1 | 1.23 | 1.26 | 1.20 | 1.33 | 1.05 |
| 2 | 1.29 | 1.26 | 1.02 | 1.22 | 0.99 |
| 3 | 1.24 | 1.37 | 1.26 | 1.52 | 1.11 |
| 5 | 1.20 | 1.14 | 0.94 | 1.08 | 1.06 |
| 7 | 0.93 | 0.94 | ---- | 1.14 | 0.92 |
| 8 | 0.98 | 0.91 | 0.93 | 1.02 | 0.90 |
| 9 | 1.02 | 1.04 | 1.05 | 1.51 | 0.96 |
| 10 | 0.98 | 0.91 | 1.30 | 1.12 | 0.87 |
| 11 | 1.10 | 0.97 | 1.26 | 1.40 | 1.12 |
| 12 | 1.13 | 0.87 | 0.98 | 1.28 | 1.25 |
| 13 | 1.03 | 1.00 | 0.45 | 1.36 | 0.99 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 76. Experiment 1--Mean serum $beta_1$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 0.30 | 0.30 | 0.25 | 0.18 | 0.29 |
| 1 | 0.29 | 0.29 | 0.30 | 0.20 | 0.27 |
| 2 | 0.34 | 0.31 | 0.34 | 0.31 | 0.32 |
| 3 | 0.28 | 0.31 | 0.26 | 0.22 | 0.34 |
| 5 | 0.40 | 0.34 | 0.47 | 0.28 | 0.27 |
| 7 | 0.37 | 0.32 | 0.37 | 0.34 | 0.32 |
| 8 | 0.58 | 0.31 | 0.65 | 0.30 | 0.39 |
| 9 | 0.28 | 0.35 | 0.42 | 0.31 | 0.30 |
| 10 | 0.35 | 0.37 | 0.32 | 0.35 | 0.35 |
| 11 | 0.30 | 0.35 | 0.51 | 0.29 | 0.25 |
| 12 | 0.31 | 0.32 | 0.40 | 0.39 | 0.27 |
| 13 | 0.32 | 0.30 | 0.36 | 0.34 | 0.31 |

[a]Prior to lead or sodium acetate exposure.

Table 77. Experiment 1--Mean serum beta$_1$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 0.34 | 0.29 | 0.37 | 0.29 | 0.41 |
| 1 | 0.33 | 0.28 | 0.29 | 0.46 | 0.40 |
| 2 | 0.38 | 0.41 | 0.34 | 0.37 | 0.37 |
| 3 | 0.28 | 0.33 | 0.38 | 0.26 | 0.36 |
| 5 | 0.35 | 0.38 | 0.48 | 0.42 | 0.40 |
| 7 | 0.39 | 0.43 | ---- | 0.44 | 0.35 |
| 8 | 0.45 | 0.42 | 0.53 | 0.46 | 0.38 |
| 9 | 0.48 | 0.37 | 0.73 | 0.42 | 0.36 |
| 10 | 0.37 | 0.54 | 0.52 | 0.55 | 0.35 |
| 11 | 0.35 | 0.37 | 0.44 | 0.43 | 0.41 |
| 12 | 0.46 | 0.43 | 0.60 | 0.57 | 0.44 |
| 13 | 0.34 | 0.34 | 0.77 | 0.46 | 0.38 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 78. Experiment 1--Mean serum beta$_2$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 0.89 | 0.91 | 0.80 | 0.84 | 0.83 |
| 1 | 0.86 | 0.90 | 0.74 | 0.71 | 1.05 |
| 2 | 0.85 | 0.82 | 0.76 | 0.84 | 0.89 |
| 3 | 0.98 | 0.88 | 0.91 | 0.80 | 0.89 |
| 5 | 0.89 | 1.01 | 0.77 | 0.87 | 1.00 |
| 7 | 0.79 | 0.89 | 0.79 | 0.87 | 0.82 |
| 8 | 0.88 | 0.82 | 0.77 | 0.86 | 0.83 |
| 9 | 0.88 | 0.86 | 0.98 | 0.98 | 1.17 |
| 10 | 0.97 | 1.06 | 0.77 | 0.79 | 1.00 |
| 11 | 1.04 | 0.79 | 1.06 | 0.92 | 1.31 |
| 12 | 1.08 | 1.44 | 1.05 | 1.14 | 1.21 |
| 13 | 1.02 | 1.05 | 0.98 | 0.88 | 1.07 |

[a]Prior to lead or sodium acetate exposure.

Table 79. Experiment 1--Mean serum $beta_2$ globulin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group | | | | |
|---|---|---|---|---|---|
| | 6 | 7 | 8[a] | 9[a] | 10 (control) |
| 0[b] | 0.91 | 0.83 | 0.88 | 0.88 | 0.88 |
| 1 | 0.86 | 0.86 | 0.93 | 0.89 | 0.80 |
| 2 | 0.96 | 0.92 | 0.87 | 0.85 | 0.83 |
| 3 | 0.84 | 0.85 | 0.94 | 0.85 | 0.82 |
| 5 | 0.87 | 0.94 | 0.94 | 0.70 | 0.98 |
| 7 | 0.88 | 0.81 | ---- | 0.83 | 0.88 |
| 8 | 0.84 | 0.95 | 0.87 | 0.72 | 0.80 |
| 9 | 0.93 | 0.84 | 1.19 | 0.99 | 1.09 |
| 10 | 0.79 | 0.79 | 1.12 | 1.00 | 0.99 |
| 11 | 0.93 | 0.94 | 1.04 | 0.79 | 1.21 |
| 12 | 1.02 | 1.05 | 0.89 | 1.12 | 1.28 |
| 13 | 0.90 | 0.87 | 1.26 | 0.82 | 1.14 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 80. Experiment 1--Mean serum gamma globulin concentrations (g/dl) of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 1.12 | 1.00 | 1.19 | 1.29 | 1.18 |
| 1 | 1.10 | 0.83 | 1.09 | 1.24 | 0.95 |
| 2 | 1.25 | 1.34 | 1.35 | 1.50 | 1.16 |
| 3 | 1.36 | 1.24 | 1.50 | 1.89 | 1.30 |
| 5 | 1.28 | 1.29 | 1.37 | 1.68 | 1.17 |
| 7 | 1.29 | 1.14 | 1.27 | 1.24 | 1.14 |
| 8 | 1.44 | 1.02 | 1.29 | 1.36 | 1.37 |
| 9 | 1.28 | 1.32 | 1.20 | 1.48 | 1.06 |
| 10 | 1.00 | 1.01 | 1.27 | 1.48 | 1.23 |
| 11 | 0.87 | 1.38 | 1.12 | 1.52 | 0.79 |
| 12 | 0.80 | 1.01 | 1.07 | 1.00 | 0.92 |
| 13 | 1.05 | 1.08 | 1.16 | 1.53 | 1.12 |

[a]Prior to lead or sodium acetate exposure.

Table 81. Experiment 1--Mean serum gamma globulin concentrations (g/dl) of groups exposed to lead or sodium acetate i.p.

| Week | Group 6 | 7 | 8[a] | 9[a] | 10 (control) |
|---|---|---|---|---|---|
| 0[b] | 1.17 | 1.40 | 1.26 | 1.15 | 1.16 |
| 1 | 1.23 | 1.34 | 1.12 | 1.18 | 1.32 |
| 2 | 1.31 | 1.54 | 1.42 | 1.33 | 1.35 |
| 3 | 1.27 | 1.55 | 1.62 | 1.44 | 1.58 |
| 5 | 1.21 | 1.69 | 1.81 | 1.90 | 1.39 |
| 7 | 1.39 | 1.89 | ---- | 1.56 | 1.24 |
| 8 | 1.41 | 2.03 | 1.88 | 1.65 | 1.36 |
| 9 | 1.28 | 1.97 | 1.89 | 1.07 | 1.26 |
| 10 | 1.02 | 1.66 | 1.98 | 1.46 | 1.21 |
| 11 | 1.37 | 2.04 | 2.11 | 2.15 | 1.15 |
| 12 | 1.18 | 1.51 | 1.61 | 1.62 | 1.12 |
| 13 | 1.31 | 1.81 | 2.02 | 1.93 | 1.14 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 82. Experiment 1--Mean serum albumin:globulin ratios of groups exposed to lead or sodium acetate orally.

| Week | Group 1 | 2 | 3 | 4 | 5 (control) |
|---|---|---|---|---|---|
| 0[a] | 0.63 | 0.61 | 0.66 | 0.52 | 0.65 |
| 1 | 0.74 | 0.81 | 0.74 | 0.67 | 0.63 |
| 2 | 0.74 | 0.70 | 0.65 | 0.53 | 0.68 |
| 3 | 0.64 | 0.70 | 0.61 | 0.50 | 0.63 |
| 5 | 0.68 | 0.65 | 0.65 | 0.57 | 0.74 |
| 7 | 0.71 | 0.90 | 0.71 | 0.62 | 0.81 |
| 8 | 0.68 | 0.96 | 0.68 | 0.72 | 0.68 |
| 9 | 0.70 | 0.74 | 0.62 | 0.55 | 0.74 |
| 10 | 0.80 | 0.94 | 0.74 | 0.69 | 0.78 |
| 11 | 0.83 | 0.81 | 0.54 | 0.58 | 0.79 |
| 12 | 0.80 | 0.64 | 0.67 | 0.64 | 0.76 |
| 13 | 0.73 | 0.76 | 0.65 | 0.59 | 0.72 |

[a]Prior to lead or sodium acetate exposure.

Table 83. Experiment 1--Mean serum albumin:globulin ratios of groups exposed to lead or sodium acetate i.p.

| Week | Group 6 | 7 | 8[a] | 9[a] | 10 (control) |
|---|---|---|---|---|---|
| 0[b] | 0.66 | 0.58 | 0.51 | 0.61 | 0.62 |
| 1 | 0.58 | 0.57 | 0.52 | 0.55 | 0.63 |
| 2 | 0.64 | 0.50 | 0.57 | 0.49 | 0.67 |
| 3 | 0.66 | 0.52 | 0.48 | 0.45 | 0.65 |
| 5 | 0.60 | 0.44 | 0.42 | 0.44 | 0.64 |
| 7 | 0.62 | 0.47 | ---- | 0.39 | 0.64 |
| 8 | 0.58 | 0.43 | 0.51 | 0.41 | 0.86 |
| 9 | 0.59 | 0.47 | 0.41 | 0.41 | 0.63 |
| 10 | 0.92 | 0.61 | 0.52 | 0.41 | 0.80 |
| 11 | 0.65 | 0.50 | 0.38 | 0.35 | 0.69 |
| 12 | 0.55 | 0.60 | 0.44 | 0.34 | 0.66 |
| 13 | 0.65 | 0.59 | 0.48 | 0.43 | 0.64 |

[a]Value for week 5 is a mean from 2 pigs, and values for weeks 7-13 are from 1 pig only.

[b]Prior to lead or sodium acetate exposure.

Table 84. Experiment 2--Mean body temperatures ($^{o}$C) during the lead or sodium acetate-exposure period prior to *Salmonella* challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| 1 | 39.8 | 39.9 | 39.9 | 39.8 | 39.4 | 39.7 |
| 3 | 40.0 | 40.0 | 39.9 | 39.9 | 39.9 | 39.6 |
| 5 | 39.4 | 39.9 | 39.3 | 39.1 | 39.5 | 39.3 |
| 8 | 39.1 | 39.4 | 39.3 | 39.4 | 39.4 | 39.3 |
| 10 | ---- | 39.6 | 39.4 | ---- | 39.4 | 39.4 |
| 12 | ---- | 39.4 | 39.7 | ---- | 39.6 | 39.4 |
| 15 | ---- | 39.5 | 39.5 | ---- | 39.1 | 39.0 |
| 17 | ---- | ---- | 39.4 | ---- | ---- | 39.0 |
| 19 | ---- | ---- | 39.2 | ---- | ---- | 38.7 |
| 21 | ---- | ---- | 39.0 | ---- | ---- | 39.2 |
| $\bar{X}$ | 39.6 | 39.7 | 39.5 | 39.6 | 39.5 | 39.3 |

Table 85. Experiment 2--Mean body temperatures ($^{o}C$) following *Salmonella* challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| 1 | 39.9 | 39.9 | 40.3 | 39.9 | 40.0 | 40.1 |
| 2 | 39.6 | 39.8 | 39.1 | 39.9 | 39.8 | 39.4 |
| 3 | 40.8 | 41.0 | 41.0 | 40.6 | 41.3 | 41.3 |
| 4 | 40.5 | 40.9 | 40.6 | 40.4 | 41.5 | 40.5 |
| 5 | 41.0 | 40.9 | 40.8 | 41.0 | 40.8 | 40.4 |
| 6 | 40.3 | 40.6 | 40.2 | 40.9 | 40.9 | 40.1 |
| 7 | 40.3 | 40.4 | 39.8 | 40.5 | 40.9 | 39.6 |
| 8 | 39.5 | 40.1 | 40.0 | 40.0 | 40.6 | 40.2 |
| 9 | 39.7 | 39.5 | 39.8 | 39.8 | 40.1 | 39.5 |
| 10 | 40.0 | 39.4 | 39.5 | 40.0 | 39.7 | 39.3 |
| $\bar{X}$ | 40.2 | 40.2 | 40.1 | 40.3 | 40.6 | 40.0 |

Table 86. Experiment 2--Mean blood lead concentrations (ug/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 4 | 5 | 4 | 9 | 5 | 6 |
| 2 | 36 | 41 | 34 | 3 | 4 | 4 |
| 4 | 39 | 38 | 35 | 1 | 1 | 2 |
| 8 | 42 | 40 | 29 | 1 | 2 | 2 |
| 15 | -- | 42 | 37 | - | 5 | 2 |
| 21 | -- | -- | 48 | - | - | 2 |
| Post-challenge | | | | | | |
| 2 | 29 | 25 | 25 | 11 | 5 | 7 |
| 9 | 17 | 16 | 18 | 3 | 3 | 2 |
| $\bar{X}$[a] | 33 | 34 | 32 | 4 | 3 | 3 |

[a]Day 0 omitted in calculating means.

Table 87. Experiment 2--Mean blood ALAD activities during lead or sodium acetate exposure and following *Salmonella* challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 228.9 | 221.0 | 219.7 | 214.5 | 222.9 | 209.7 |
| 2 | 56.2 | 55.0 | 48.6 | 208.2 | 223.2 | 194.2 |
| 4 | 37.9 | 41.0 | 39.5 | 210.3 | 234.2 | 203.0 |
| 8 | 59.8 | 59.0 | 58.2 | 246.2 | 252.6 | 226.6 |
| 15 | ---- | 38.8 | 35.2 | ----- | 227.3 | 196.9 |
| 21 | ---- | ---- | 14.5 | ----- | ----- | 152.3 |
| Post-challenge | | | | | | |
| 2 | 69.7 | 42.8 | 16.7 | 216.2 | 220.7 | 131.7 |
| 9 | 35.4 | 25.3 | 16.0 | 126.2 | 180.8 | 129.6 |
| $\bar{X}^a$ | 51.8 | 43.6 | 32.7 | 201.4 | 223.1 | 176.3 |

[a]Day 0 omitted in calculating means.

Table 88. Experiment 2--Mean hematocrit percentages during lead or sodium acetate exposure and following *Salmonella* challenge.

| Day | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 33 | 34 | 32 | 34 | 35 | 33 |
| 8 | 33 | 33 | 32 | 31 | 32 | 32 |
| 15 | -- | 35 | 34 | -- | 33 | 32 |
| 21 | -- | -- | 33 | -- | -- | 33 |
| $\bar{X}$[a] | 33 | 34 | 33 | 31 | 32 | 32 |
| Post-challenge | | | | | | |
| 2 | 34 | 33 | 32 | 31 | 32 | 33 |
| 4 | 28 | 33 | 32 | 28 | 31 | 32 |
| 9 | 26 | 29 | 30 | 25 | 28 | 29 |
| $\bar{X}$ | 29 | 32 | 31 | 28 | 30 | 31 |

[a]Day 0 omitted in calculating means.

Table 89. Experiment 2--Mean total leukocyte counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Days | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 15,917 | 15,250 | 17,650 | 13,967 | 17,117 | 19,717 |
| 8 | 16,033 | 17,000 | 18,417 | 19,783 | 18,633 | 22,033 |
| 15 | ------ | 17,633 | 19,900 | ------ | 19,283 | 22,650 |
| 21 | ------ | ------ | 20,267 | ------ | ------ | 22,550 |
| $\bar{X}$[a] | 16,033 | 17,316 | 19,528 | 19,783 | 18,958 | 22,411 |
| Post-challenge | | | | | | |
| 2 | 27,033 | 20,167 | 28,033 | 33,017 | 25,783 | 27,000 |
| 4 | 22,817 | 16,200 | 21,650 | 20,767 | 19,300 | 17,917 |
| 9 | 20,633 | 16,967 | 22,100 | 18,883 | 19,240 | 21,317 |
| $\bar{X}$ | 23,494 | 17,778 | 23,928 | 24,222 | 21,441 | 22,078 |

[a]Day 0 omitted in calculating means.

Table 90. Experiment 2--Mean total segmented neutrophil counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 5,220 | 5,614 | 7,016 | 5,182 | 6,158 | 8,068 |
| 8 | 4,032 | 4,464 | 4,065 | 7,117 | 6,098 | 8,487 |
| 15 | ----- | 6,638 | 4,800 | ----- | 7,210 | 9,275 |
| 21 | ----- | ----- | 6,114 | ----- | ----- | 7,775 |
| $\bar{X}$[a] | 4,032 | 5,551 | 4,993 | 7,117 | 6,654 | 8,512 |
| Post-challenge | | | | | | |
| 2 | 9,558 | 2,179 | 14,265 | 11,434 | 4,024 | 10,772 |
| 4 | 5,711 | 3,220 | 6,329 | 3,355 | 3,992 | 3,203 |
| 9 | 5,781 | 5,527 | 7,001 | 5,831 | 5,459 | 7,609 |
| $\bar{X}$ | 7,017 | 3,642 | 9,198 | 6,873 | 4,492 | 7,195 |

[a]Day 0 omitted in calculating means.

Table 91. Experiment 2--Mean total band neutrophil counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 27 | 27 | 161 | 140 | 105 | 57 |
| 8 | 210 | 27 | 281 | 89 | 160 | 110 |
| 15 | --- | 288 | 848 | -- | 916 | 790 |
| 21 | --- | --- | 1,783 | -- | --- | 1,836 |
| $\bar{X}$[a] | 210 | 158 | 971 | 89 | 538 | 912 |
| Post-challenge | | | | | | |
| 2 | 8,394 | 10,354 | 5,589 | 9,498 | 13,353 | 10,412 |
| 4 | 8,181 | 5,080 | 6,066 | 7,238 | 6,446 | 5,809 |
| 9 | 4,317 | 3,516 | 7,532 | 5,215 | 5,895 | 9,564 |
| $\bar{X}$ | 6,964 | 6,317 | 6,396 | 7,317 | 8,565 | 8,595 |

[a]Day 0 omitted in calculating means.

Table 92. Experiment 2--Mean total lymphocyte counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposure) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 10,136 | 9,223 | 10,087 | 8,221 | 10,402 | 11,340 |
| 8 | 11,423 | 12,222 | 13,102 | 12,266 | 11,826 | 13,112 |
| 15 | ------ | 10,329 | 13,547 | ------ | 10,510 | 12,106 |
| 21 | ------ | ------ | 11,832 | ------ | ------ | 12,697 |
| $\bar{X}$[a] | 11,423 | 11,276 | 12,827 | 12,266 | 11,168 | 12,638 |
| Post-challenge | | | | | | |
| 2 | 8,392 | 7,603 | 8,180 | 11,290 | 8,075 | 5,778 |
| 4 | 8,638 | 7,479 | 8,797 | 9,735 | 8,196 | 8,834 |
| 9 | 9,985 | 7,555 | 7,219 | 7,405 | 7,469 | 3,937 |
| $\bar{X}$ | 9,005 | 7,546 | 8,065 | 9,477 | 7,913 | 6,183 |

[a]Day 0 omitted in calculating means.

Table 93. Experiment 2--Mean total eosinophil counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 390 | 248 | 243 | 323 | 364 | 94 |
| 8 | 267 | 215 | 590 | 186 | 370 | 179 |
| 15 | --- | 342 | 705 | --- | 584 | 479 |
| 21 | --- | --- | 508 | --- | --- | 335 |
| $\bar{X}$[a] | 267 | 278 | 601 | 186 | 477 | 331 |
| Post-challenge | | | | | | |
| 2 | 173 | 30 | 0 | 164 | 330 | 0 |
| 4 | 167 | 337 | 458 | 314 | 546 | 71 |
| 9 | 346 | 370 | 348 | 393 | 454 | 207 |
| $\bar{X}$ | 229 | 246 | 403 | 290 | 443 | 93 |

[a]Day 0 omitted in calculating means.

Table 94. Experiment 2--Mean total monocyte counts (per cubic mm) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 143 | 138 | 143 | 125 | 87 | 231 |
| 8 | 102 | 272 | 443 | 124 | 120 | 146 |
| 15 | --- | 37 | 0 | --- | 28 | 0 |
| 21 | --- | -- | 29 | --- | -- | 0 |
| $\bar{X}$[a] | 102 | 154 | 157 | 124 | 74 | 49 |
| Post-challenge | | | | | | |
| 2 | 516 | 0 | 0 | 631 | 0 | 38 |
| 4 | 154 | 85 | 0 | 125 | 65 | 0 |
| 9 | 43 | 0 | 0 | 0 | 0 | 0 |
| $\bar{X}$ | 238 | 28 | 0 | 252 | 22 | 13 |

[a]Day 0 omitted in calculating means.

Table 95. Experiment 2--Mean plasma protein:fibrinogen ratios during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 20 | 16 | 16 | 16 | 18 | 15 |
| 8 | 24 | 26 | 26 | 29 | 22 | 24 |
| 15 | -- | 22 | 25 | -- | 18 | 21 |
| 21 | -- | -- | 27 | -- | -- | 21 |
| $\bar{X}$[a] | 24 | 24 | 26 | 29 | 20 | 22 |
| Post-challenge | | | | | | |
| 2 | 15 | 11 | 14 | 11 | 11 | 12 |
| 4 | 11 | 42 | 14 | 10 | 52 | 16 |
| 9 | 13 | 15 | 14 | 11 | 17 | 15 |
| $\bar{X}$ | 13 | 23 | 14 | 11 | 27 | 14 |

[a]Day 0 omitted in calculating means.

Table 96. Experiment 2--Mean total serum protein concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 5.5 | 6.0 | 5.8 | 5.4 | 5.7 | 5.8 |
| 2 | 5.6 | 6.2 | 6.0 | 5.6 | 6.2 | 6.0 |
| 4 | 6.1 | 6.1 | 6.0 | 5.5 | 5.8 | 6.0 |
| 8 | 6.4 | 6.5 | 6.4 | 5.9 | 6.2 | 6.1 |
| 15 | --- | 6.6 | 6.5 | --- | 6.3 | 6.4 |
| 21 | --- | --- | 6.4 | --- | --- | 6.4 |
| $\bar{X}$[a] | 6.0 | 6.4 | 6.3 | 5.7 | 6.1 | 6.2 |
| Post-challenge | | | | | | |
| 2 | 6.2 | 7.2 | 6.8 | 6.1 | 6.9 | 7.0 |
| 4 | 6.2 | 7.3 | 6.9 | 6.4 | 6.9 | 7.0 |
| 7 | 6.3 | 6.8 | 7.1 | 6.8 | 6.6 | 7.2 |
| 9 | 6.7 | 6.6 | 6.8 | 6.6 | 6.9 | 7.1 |
| $\bar{X}$ | 6.4 | 7.0 | 6.9 | 6.5 | 6.8 | 7.1 |

[a]Day 0 omitted in calculating means.

Table 97. Experiment 2--Mean serum albumin concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 2.02 | 2.24 | 2.17 | 2.06 | 2.10 | 2.11 |
| 2 | 2.05 | 2.24 | 2.16 | 2.15 | 2.25 | 2.18 |
| 4 | 2.26 | 2.26 | 2.17 | 2.07 | 2.12 | 2.20 |
| 8 | 2.27 | 2.34 | 2.28 | 2.13 | 2.17 | 2.06 |
| 15 | ---- | 2.51 | 2.24 | ---- | 2.22 | 2.13 |
| 21 | ---- | ---- | 2.18 | ---- | ---- | 2.21 |
| $\bar{X}$[a] | 2.19 | 2.34 | 2.21 | 2.11 | 2.19 | 2.16 |
| Post-challenge | | | | | | |
| 2 | 2.09 | 2.23 | 2.13 | 1.88 | 2.09 | 2.09 |
| 4 | 1.96 | 2.20 | 2.00 | 1.87 | 1.98 | 1.99 |
| 7 | 1.87 | 2.10 | 1.99 | 1.87 | 1.90 | 2.03 |
| 9 | 2.01 | 1.96 | 1.90 | 1.94 | 2.07 | 1.96 |
| $\bar{X}$ | 1.98 | 2.12 | 2.00 | 1.89 | 2.01 | 2.02 |

[a]Day 0 omitted in calculating means.

Table 98. Experiment 2--Mean serum alpha$_1$ globulin concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 0.30 | 0.36 | 0.35 | 0.34 | 0.37 | 0.38 |
| 2 | 0.31 | 0.34 | 0.40 | 0.31 | 0.36 | 0.37 |
| 4 | 0.32 | 0.29 | 0.38 | 0.31 | 0.30 | 0.37 |
| 8 | 0.33 | 0.34 | 0.36 | 0.31 | 0.30 | 0.37 |
| 15 | ---- | 0.28 | 0.37 | ---- | 0.32 | 0.40 |
| 21 | ---- | ---- | 0.34 | ---- | ---- | 0.35 |
| $\bar{X}$[a] | 0.33 | 0.31 | 0.37 | 0.31 | 0.32 | 0.37 |
| Post-challenge | | | | | | |
| 2 | 0.29 | 0.37 | 0.35 | 0.41 | 0.36 | 0.41 |
| 4 | 0.30 | 0.37 | 0.38 | 0.37 | 0.30 | 0.39 |
| 7 | 0.29 | 0.30 | 0.35 | 0.31 | 0.22 | 0.39 |
| 9 | 0.24 | 0.27 | 0.40 | 0.30 | 0.27 | 0.35 |
| $\bar{X}$ | 0.28 | 0.33 | 0.37 | 0.35 | 0.29 | 0.38 |

[a]Day 0 omitted in calculating means.

Table 99. Experiment 2--Mean serum alpha$_2$ globulin concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 1.19 | 1.23 | 1.21 | 1.08 | 1.16 | 1.16 |
| 2 | 1.21 | 1.30 | 1.22 | 1.15 | 1.28 | 1.20 |
| 4 | 1.28 | 1.24 | 1.19 | 1.12 | 1.17 | 1.20 |
| 8 | 1.35 | 1.31 | 1.21 | 1.23 | 1.21 | 1.26 |
| 15 | ---- | 1.13 | 1.10 | ---- | 1.16 | 1.19 |
| 21 | ---- | ---- | 1.20 | ---- | ---- | 1.14 |
| $\bar{X}$[a] | 1.28 | 1.24 | 1.19 | 1.16 | 1.21 | 1.20 |
| Post-challenge | | | | | | |
| 2 | 1.44 | 1.66 | 1.47 | 1.36 | 1.63 | 1.56 |
| 4 | 1.55 | 1.73 | 1.66 | 1.70 | 1.80 | 1.74 |
| 7 | 1.64 | 1.63 | 1.75 | 1.90 | 1.77 | 1.75 |
| 9 | 1.69 | 1.61 | 1.49 | 1.81 | 1.76 | 1.55 |
| $\bar{X}$ | 1.58 | 1.66 | 1.59 | 1.69 | 1.74 | 1.65 |

[a]Day 0 omitted in calculating means.

Table 100. Experiment 2--Mean serum $beta_1$ globulin concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| Day | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 0.42 | 0.46 | 0.43 | 0.42 | 0.40 | 0.42 |
| 2 | 0.43 | 0.47 | 0.45 | 0.38 | 0.46 | 0.42 |
| 4 | 0.48 | 0.45 | 0.49 | 0.38 | 0.44 | 0.40 |
| 8 | 0.46 | 0.49 | 0.57 | 0.42 | 0.48 | 0.50 |
| 15 | ---- | 0.43 | 0.52 | ---- | 0.43 | 0.49 |
| 21 | ---- | ---- | 0.55 | ---- | ---- | 0.44 |
| $\bar{X}$[a] | 0.46 | 0.46 | 0.52 | 0.39 | 0.45 | 0.45 |
| Post-challenge | | | | | | |
| 2 | 0.51 | 0.60 | 0.64 | 0.53 | 0.54 | 0.62 |
| 4 | 0.55 | 0.74 | 0.71 | 0.65 | 0.70 | 0.70 |
| 7 | 0.58 | 0.60 | 0.70 | 0.80 | 0.66 | 0.70 |
| 9 | 0.68 | 0.58 | 0.72 | 0.65 | 0.68 | 0.82 |
| $\bar{X}$ | 0.58 | 0.63 | 0.69 | 0.66 | 0.64 | 0.71 |

[a]Day 0 omitted in calculating means.

Table 101. Experiment 2--Mean serum $beta_2$ globulin concentrations (g/dl) during lead or sodium acetate exposure and following Salmonella challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 0.84 | 0.86 | 0.86 | 0.75 | 0.80 | 0.92 |
| 2 | 0.85 | 0.86 | 0.94 | 0.71 | 0.85 | 0.92 |
| 4 | 0.87 | 0.89 | 0.88 | 0.76 | 0.80 | 0.87 |
| 8 | 0.97 | 0.93 | 0.90 | 0.79 | 0.82 | 0.87 |
| 15 | ---- | 0.96 | 0.98 | ---- | 0.91 | 0.91 |
| 21 | ---- | ---- | 0.94 | ---- | ---- | 0.98 |
| $\bar{x}$[a] | 0.90 | 0.91 | 0.93 | 0.75 | 0.84 | 0.91 |
| Post-challenge | | | | | | |
| 2 | 0.81 | 0.98 | 0.95 | 0.86 | 0.92 | 0.99 |
| 4 | 0.84 | 0.92 | 0.91 | 0.78 | 0.81 | 0.90 |
| 7 | 0.83 | 0.76 | 0.87 | 0.78 | 0.72 | 0.87 |
| 9 | 0.82 | 0.76 | 0.83 | 0.77 | 0.76 | 0.83 |
| $\bar{X}$ | 0.82 | 0.86 | 0.89 | 0.80 | 0.76 | 0.90 |

[a]Day 0 omitted in calculating means.

Table 102. Experiment 2--Mean serum gamma globulin concentrations (g/dl) during lead or sodium acetate exposure and following *Salmonella* challenge.

| | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| Day | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 0.75 | 0.86 | 0.78 | 0.77 | 0.90 | 0.79 |
| 2 | 0.78 | 0.92 | 0.84 | 0.87 | 0.99 | 0.86 |
| 4 | 0.86 | 0.97 | 0.88 | 0.87 | 1.02 | 0.99 |
| 8 | 1.02 | 1.12 | 1.08 | 0.99 | 1.17 | 1.05 |
| 15 | ---- | 1.28 | 1.25 | ---- | 1.27 | 1.31 |
| 21 | ---- | ---- | 1.23 | ---- | ---- | 1.29 |
| $\bar{x}$[a] | 0.89 | 1.07 | 1.06 | 0.91 | 1.11 | 1.10 |
| Post-challenge | | | | | | |
| 2 | 1.10 | 1.34 | 1.22 | 1.09 | 1.35 | 1.30 |
| 4 | 1.02 | 1.36 | 1.26 | 1.06 | 1.31 | 1.25 |
| 7 | 1.14 | 1.40 | 1.39 | 1.00 | 1.31 | 1.43 |
| 9 | 1.28 | 1.44 | 1.48 | 1.17 | 1.40 | 1.56 |
| $\bar{X}$ | 1.14 | 1.38 | 1.34 | 1.08 | 1.34 | 1.39 |

[a]Day 0 omitted in calculating means.

Table 103. Experiment 2--Mean serum albumin:globulin ratios during lead or sodium acetate exposure and following Salmonella challenge.

| Day | Group A (lead-exposed) | | | Group B (control) | | |
|---|---|---|---|---|---|---|
| | Rep 1 | Rep 2 | Rep 3 | Rep 1 | Rep 2 | Rep 3 |
| Exposure | | | | | | |
| 0 | 0.58 | 0.60 | 0.60 | 0.62 | 0.58 | 0.57 |
| 2 | 0.57 | 0.58 | 0.56 | 0.64 | 0.57 | 0.58 |
| 4 | 0.59 | 0.59 | 0.57 | 0.60 | 0.57 | 0.58 |
| 8 | 0.55 | 0.56 | 0.55 | 0.57 | 0.54 | 0.51 |
| 15 | ---- | 0.61 | 0.53 | ---- | 0.54 | 0.50 |
| 21 | ---- | ---- | 0.51 | ---- | ---- | 0.53 |
| $\bar{X}$[a] | 0.57 | 0.58 | 0.54 | 0.60 | 0.56 | 0.54 |
| Post-challenge | | | | | | |
| 2 | 0.50 | 0.45 | 0.46 | 0.44 | 0.44 | 0.43 |
| 4 | 0.46 | 0.43 | 0.41 | 0.42 | 0.40 | 0.40 |
| 7 | 0.36 | 0.44 | 0.40 | 0.34 | 0.41 | 0.40 |
| 9 | 0.43 | 0.42 | 0.39 | 0.41 | 0.43 | 0.38 |
| $\bar{X}$ | 0.44 | 0.44 | 0.42 | 0.40 | 0.42 | 0.40 |

[a]Day 0 omitted in calculating means.

Table 104. Experiment 3--Mean body temperatures ($^{o}C$) during the lead or sodium chloride-exposure period prior to Salmonella challenge (day 15).

| Day | Group | | |
|---|---|---|---|
| | A (control) | B | C |
| 1 | 39.2 | 39.1 | 39.0 |
| 2 | 39.1 | 39.6 | 39.3 |
| 3 | 39.2 | 38.9 | 39.6 |
| 4 | 39.4 | 39.3 | 39.5 |
| 5 | 39.5 | 39.4 | 39.7 |
| 6 | 39.5 | 39.3 | 40.0 |
| 7 | 39.6 | 39.8 | 39.4 |
| 8 | 39.7 | 39.8 | 39.8 |
| 9 | 39.9 | 39.8 | 39.7 |
| 10 | 39.8 | 39.8 | 39.6 |
| 11 | 39.7 | 39.6 | 39.1 |
| 12 | 39.8 | 39.4 | 39.1 |
| 13 | 39.7 | 39.3 | 39.2 |
| 14 | 40.2 | 39.3 | 39.1 |
| 15 | 39.7 | 39.3 | 38.8 |

Table 105. Experiment 3--Mean body temperatures ($^{o}C$) following Salmonella challenge (day 15).

| Day | Group | | |
|---|---|---|---|
| | A (control) | B | C |
| 16 | 39.7 | 39.1 | 38.7 |
| 17 | 39.9 | 39.3 | 38.7 |
| 18 | 40.6 | 41.1 | 40.7 |
| 19 | 40.4 | 40.9 | 41.1 |
| 20 | 39.8 | 40.2 | 40.3 |
| 21 | 40.1 | 40.1 | 40.7 |
| 22 | 39.8 | 40.4 | 40.0 |
| 23 | 40.2 | 41.2 | 40.2 |
| 24 | 40.1 | 40.9 | 39.9 |
| 25 | 39.3 | 41.2 | 39.8 |
| 26 | 39.6 | 39.5 | 40.0 |
| 27 | 39.3 | 40.0 | 39.9 |
| 28 | 38.9 | ---- | 39.9 |
| 29 | 39.6 | ---- | 39.0 |

Table 106. Experiment 3--Mean blood lead concentrations (ug/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| 1 | 2[a] | 2[a] | -- |
| 6 | 3 | 98 | -- |
| 8 | 12 | 118 | 9[a] |
| 13 | 3 | 107 | 92 |
| 15 | 1 | 97 | 86 |
| 21 | 5 | 26 | 24 |

[a]Prior to lead or sodium chloride exposure.

Table 107. Experiment 3--Mean blood ALAD activities during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| 1 | 109.8[a] | 112.8[a] | No collection |
| 6 | 129.9 | 20.2 | No collection |
| 8 | 124.2 | 16.4 | 127.1[a] |
| 13 | 136.4 | 14.9 | 23.9 |
| 15 | 135.6 | 17.1 | 25.7 |
| 21 | 97.2 | 9.0 | 14.2 |

[a]Prior to lead or sodium chloride exposure.

Table 108. Experiment 3--Mean blood hemoglobin concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Lead Expos. | | | |
| 1 | 10.6[a] | 10.8[a] | 10.3[a] |
| 6 | 9.2 | 9.5 | 8.8[a] |
| 8 | 8.3 | 8.3 | 8.4 |
| 13 | 8.7 | 8.1 | 7.8 |
| 15 | 8.0 | 7.7 | 7.3 |
| Post-Inoc. | | | |
| 17 | 8.5 | 8.3 | 7.8 |
| 18 | 7.9 | 7.8 | 7.3 |
| 20 | 8.3 | 7.8 | 6.8 |
| 22 | 8.4 | 6.8 | 7.2 |
| 24 | 8.3 | 6.9 | 7.1 |

[a]Prior to lead or sodium chloride exposure.

Table 109. Experiment 3--Mean hematocrit percentages during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Lead Expos. | | | |
| 1 | 33[a] | 34[a] | 33[a] |
| 6 | 29 | 31 | 29[a] |
| 8 | 27 | 29 | 27 |
| 13 | 28 | 28 | 26 |
| 15 | 27 | 26 | 26 |
| Post-Inoc. | | | |
| 17 | 27 | 28 | 27 |
| 18 | 27 | 29 | 26 |
| 20 | 29 | 27 | 24 |
| 22 | 29 | 25 | 26 |
| 24 | 30 | 25 | 26 |

[a]Prior to lead or sodium chloride exposure.

Table 110. Experiment 3--Mean total red blood cell counts x $10^6$ (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 4.8[a] | 5.1[a] | 5.2[a] |
| 6 | 5.6 | 6.0 | 6.0[a] |
| 8 | 5.0 | 4.8 | 5.0 |
| 13 | 4.6 | 5.4 | 4.8 |
| 15 | 4.8 | 4.9 | 4.7 |
| Post-challenge | | | |
| 17 | 5.4 | 5.8 | 5.6 |
| 18 | 4.3 | 5.2 | 5.1 |
| 20 | 5.2 | 5.1 | 4.6 |
| 22 | 5.3 | 4.6 | 4.8 |
| 24 | 5.5 | 4.8 | 4.8 |

[a]Prior to lead or sodium chloride exposure.

Table 111. Experiment 3--Mean MCVs (fl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 68.5[a] | 66.8[a] | 63.5[a] |
| 6 | 51.4 | 51.5 | 48.0[a] |
| 8 | 55.5 | 61.3 | 54.8 |
| 13 | 61.4 | 51.5 | 55.8 |
| 15 | 58.0 | 53.7 | 54.5 |
| Post-challenge | | | |
| 17 | 50.7 | 50.0 | 49.0 |
| 18 | 63.0 | 55.5 | 51.1 |
| 20 | 55.5 | 53.2 | 53.1 |
| 22 | 54.9 | 55.2 | 53.7 |
| 24 | 53.9 | 53.5 | 53.6 |

[a]Prior to lead or sodium chloride exposure.

Table 112. Experiment 3--Mean MCHCs (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 32.1[a] | 31.8[a] | 31.4[a] |
| 6 | 32.2 | 30.5 | 30.8[a] |
| 8 | 30.9 | 28.5 | 30.6 |
| 13 | 30.8 | 29.4 | 29.6 |
| 15 | 30.2 | 29.4 | 28.2 |
| Post-challenge | | | |
| 17 | 30.8 | 30.0 | 29.1 |
| 18 | 29.3 | 27.4 | 28.2 |
| 20 | 29.2 | 28.5 | 28.3 |
| 22 | 28.6 | 27.1 | 27.9 |
| 24 | 28.1 | 27.7 | 27.3 |

[a]Prior to lead or sodium chloride exposure.

Table 113. Experiment 3--Mean MCHs (pg) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 22.1[a] | 21.2[a] | 19.6[a] |
| 6 | 16.6 | 15.7 | 14.5 |
| 8 | 16.9 | 17.2 | 16.6 |
| 13 | 18.7 | 15.0 | 16.4 |
| 15 | 17.0 | 15.7 | 15.5 |
| Post-challenge | | | |
| 17 | 15.7 | 14.4 | 14.2 |
| 18 | 18.4 | 15.2 | 14.3 |
| 20 | 16.0 | 15.1 | 15.0 |
| 22 | 15.6 | 14.8 | 15.0 |
| 24 | 15.0 | 14.2 | 14.6 |

[a]Prior to lead or sodium chloride exposure.

Table 114. Experiment 3--Mean total leukocyte counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 17,750[a] | 16,500[a] | 17,825[a] |
| 6 | 20,300 | 22,575 | 21,876[a] |
| 8 | 16,025 | 18,288 | 17,175 |
| 13 | 20,375 | 24,125 | 22,288 |
| 15 | 21,838 | 21,688 | 17,925 |
| Post-challenge | | | |
| 17 | 30,062 | 26,888 | 24,588 |
| 18 | 24,800 | 30,612 | 24,438 |
| 20 | 21,738 | 17,557 | 13,062 |
| 22 | 30,033 | 20,100 | 18,243 |
| 24 | 36,280 | 21,467 | 24,533 |

[a]Prior to lead or sodium chloride exposure.

Table 115. Experiment 3--Mean total segmented neutrophil counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 9,757[a] | 10,712[a] | 8,069[a] |
| 6 | 11,472 | 12,876 | 12,675[a] |
| 8 | 8,315 | 9,514 | 9,550 |
| 13 | 11,332 | 11,363 | 11,738 |
| 15 | 12,407 | 11,097 | 9,833 |
| Post-challenge | | | |
| 17 | 15,681 | 14,310 | 10,270 |
| 18 | 5,644 | 10,776 | 9,704 |
| 20 | 4,272 | 1,138 | 878 |
| 22 | 8,390 | 1,825 | 2,192 |
| 24 | 14,093 | 1,498 | 4,989 |

[a]Prior to lead or sodium chloride exposure.

Table 116. Experiment 3--Mean total band neutrophil counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 1,003[a] | 1,291[a] | 828[a] |
| 6 | 836 | 571 | 751[a] |
| 8 | 425 | 1,212 | 454 |
| 13 | 445 | 0 | 192 |
| 15 | 232 | 27 | 0 |
| Post-challenge | | | |
| 17 | 4,105 | 623 | 286 |
| 18 | 10,602 | 13,992 | 8,050 |
| 20 | 8,679 | 9,810 | 6,407 |
| 22 | 8,383 | 13,320 | 9,110 |
| 24 | 8,871 | 14,623 | 13,070 |

[a]Prior to lead or sodium chloride exposure.

Table 117. Experiment 3--Mean total lymphocyte counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 6,335[a] | 4,337[a] | 8,639[a] |
| 6 | 7,702 | 8,025 | 7,855[a] |
| 8 | 7,399 | 7,119 | 6,785 |
| 13 | 8,422 | 12,518 | 10,140 |
| 15 | 8,951 | 10,132 | 7,915 |
| Post-challenge | | | |
| 17 | 10,070 | 11,212 | 13,428 |
| 18 | 8,173 | 5,510 | 6,519 |
| 20 | 8,253 | 6,552 | 5,627 |
| 22 | 12,451 | 4,895 | 6,674 |
| 24 | 12,914 | 4,838 | 6,315 |

[a]Prior to lead or sodium chloride exposure.

Table 118. Experiment 3--Mean total eosinophil counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 281[a] | 55[a] | 112[a] |
| 6 | 264 | 746 | 197[a] |
| 8 | 250 | 333 | 296 |
| 13 | 175 | 243 | 153 |
| 15 | 248 | 431 | 176 |
| Post-challenge | | | |
| 17 | 282 | 489 | 461 |
| 18 | 122 | 0 | 18 |
| 20 | 206 | 0 | 134 |
| 22 | 456 | 0 | 177 |
| 24 | 340 | 0 | 87 |

[a]Prior to lead or sodium chloride exposure.

Table 119. Experiment 3--Mean total monocyte counts (per cubic mm) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 1 | 329[a] | 84[a] | 196[a] |
| 6 | 0 | 197 | 269[a] |
| 8 | 14 | 131 | 77 |
| 13 | 0 | 0 | 0 |
| 15 | 0 | 0 | 0 |
| Post-challenge | | | |
| 17 | 448 | 264 | 28 |
| 18 | 258 | 0 | 168 |
| 20 | 286 | 78 | 0 |
| 22 | 352 | 120 | 89 |
| 24 | 63 | 449 | 73 |

[a]Prior to lead or sodium chloride exposure.

Table 120. Experiment 3--Mean plasma protein:fibrinogen ratios during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | B | C |
|---|---|---|---|
| Exposure | | | |
| 1 | 40.3[a] | 47.0[a] | 38.6[a] |
| 6 | 28.8 | 28.1 | 28.5[a] |
| 8 | 27.3 | 39.2 | 19.5 |
| 13 | 26.0 | 32.3 | 30.0 |
| 15 | 18.4 | 39.5 | 35.3 |
| Post-challenge | | | |
| 17 | 20.0 | 38.7 | 41.6 |
| 18 | 16.9 | 34.7 | 33.9 |
| 20 | 28.4 | 28.0 | 21.0 |
| 22 | 17.1 | 14.0 | 10.0 |
| 24 | 22.8 | 14.9 | 15.7 |

[a]Prior to lead or sodium chloride exposure.

Table 121. Experiment 3--Mean total serum protein concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | B | C |
|---|---|---|---|
| Exposure | | | |
| 6 | 4.4 | 4.6 | 4.4 |
| 13 | 4.8 | 4.2 | 4.4 |
| Post-challenge | | | |
| 17 | 5.5 | 5.2 | 5.2 |
| 20 | 5.3 | 4.8 | 4.6 |
| 22 | 5.2 | 4.6 | 4.4 |

Table 122. Experiment 3--Mean serum albumin concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group | | |
|---|---|---|---|
| | A (control) | B | C |
| Exposure | | | |
| 6 | 1.42 | 1.60 | 1.46 |
| 13 | 1.44 | 1.38 | 1.46 |
| Post-challenge | | | |
| 17 | 1.61 | 1.74 | 1.78 |
| 20 | 1.34 | 1.35 | 1.42 |
| 22 | 1.39 | 1.28 | 1.16 |

Table 123. Experiment 3--Mean serum alpha globulin concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 6 | 1.60 | 1.55 | 1.62 |
| 13 | 1.68 | 1.28 | 1.45 |
| Post-challenge | | | |
| 17 | 2.05 | 1.58 | 1.58 |
| 20 | 2.24 | 1.63 | 1.57 |
| 22 | 2.03 | 1.65 | 1.65 |

Table 124. Experiment 3--Mean serum beta globulin concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group | | |
|---|---|---|---|
| | A (control) | B | C |
| Exposure | | | |
| 6 | 0.70 | 0.79 | 0.76 |
| 13 | 0.83 | 0.73 | 0.76 |
| Post-challenge | | | |
| 17 | 0.88 | 0.89 | 0.82 |
| 20 | 0.84 | 0.82 | 0.81 |
| 22 | 0.87 | 0.77 | 0.74 |

Table 125. Experiment 3--Mean serum gamma globulin concentrations (g/dl) during lead or sodium chloride exposure and following Salmonella challenge (day 15).

| Day | Group A (control) | Group B | Group C |
|---|---|---|---|
| Exposure | | | |
| 6 | 0.66 | 0.68 | 0.62 |
| 13 | 0.86 | 0.78 | 0.78 |
| Post-challenge | | | |
| 17 | 0.95 | 0.90 | 0.92 |
| 20 | 0.85 | 0.97 | 0.86 |
| 22 | 0.89 | 0.93 | 0.76 |

Table 126. Experiment 3--Mean albumin:globulin ratios during lead or sodium chloride exposure and following *Salmonella* challenge (day 15).

| Day | Group | | |
|---|---|---|---|
| | A (control) | B | C |
| *Exposure* | | | |
| 6 | 0.48 | 0.53 | 0.49 |
| 13 | 0.43 | 0.49 | 0.49 |
| *Post-challenge* | | | |
| 17 | 0.41 | 0.51 | 0.54 |
| 20 | 0.34 | 0.40 | 0.44 |
| 22 | 0.37 | 0.38 | 0.37 |