EXPERIMENTAL VIBRIONIC COLITIS IN SWINE

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ABSTRACT

EXPERIMENTAL VIBRIONIC COLITIS IN SWINE

by George R. Ruth

A total of 16, 5-week-old Yorkshire pigs was used to study the pathology and changes in serum electrolyte and hemogram values due to an infectious colitis caused by Vibrio coli.

The clinical signs and gross and histologic lesions experimentally produced were typical of those noted for naturally occurring field cases. The clinical signs consisted of dehydration, anorexia, cyanosis, dysentery, prostration, and death. The lesions were primarily a necrotic-hemorrhagic colitis and typhlitis.

As the dysentery progressed, hematocrit and hemoglobin increased significantly, while blood pH and blood volume decreased. Serum sodium, chloride, and bicarbonate declined on the 3rd day of the dysentery; potassium and phosphorus increased at prostration. Calcium and magnesium did not vary markedly from the controls.

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By

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INTRODUCTION

Research was conducted at the Michigan State University Veterinary Clinic in the fall of 1967 to obtain additional basic information on the disease swine dysentery, which is an infectious colitis. Young pigs were used as experimental animals.

The disease was produced using the same infectious material and general techniques as employed by the Eli Lilly Veterinary Research Laboratories. In these laboratories, rather extensive research has been conducted in experimentally produced swine dysentery for evaluation of drug efficacy in treatment and prevention of the disease. The infectious material obtained for Lilly's research was supplied by veterinarians from established field cases of swine dysentery. This material, believed to contain no other pathogens, was used in their laboratory to produce the disease. A spiral colon taken from such an experimentally diseased pig was used as the inoculum for this research.

OBJECTIVES

The objectives of this research were:

1. To observe and record the clinical signs of experimentally produced swine dysentery.

2. To investigate the influence of an experimentally produced infectious colitis in young swine on the following blood factors:

   a. Serum electrolytes

      1). Sodium
      2). Potassium
      3). Chloride
      4). Bicarbonate
      5). Inorganic phosphorus
      6). Magnesium
      7). Calcium

   b. pH
   c. Hemogram
   d. Volume

3. To study the gross and microscopic pathology of experimentally produced swine dysentery.

4. To evaluate swine dysentery as an experimental model in order to obtain basic information on electrolyte metabolism during colitis for comparative pathologic and medical purposes.

REVIEW OF LITERATURE

Whiting and Doyle (1921), at Purdue University, first reported the disease syndrome called swine dysentery. Since that time, many investigators throughout the world have described the clinical signs and pathologic lesions of the disease. Though many of these reports described the general signs, incidence, and treatments that were believed to be effective at that time, the diagnosis might be questioned in several cases. Therefore, this review is limited primarily to the more significant studies on the disease.*

Description of the Disease

Clinical signs. Lussier (1961) made a detailed study of 249 outbreaks of swine dysentery in Canada and reported that it is a peracute, acute, subacute, or chronic disease. Hofferd (1936) stated that a minimum incubation time of 2 days could occur with stress, while Doyle (1939), in Indiana, recorded a maximum period of 60 days. Roberts (1956), reporting from Australia, mentioned the average incubation time to be about 2 weeks.

The disease appears in any season (Lussier, 1961) and may have any or all of the following signs: depression, excessive thirst, limp tail, and mucus in dark stools (Deas, 1960); arched back and sunken eyes (Deas,

*A more complete listing of foreign references is given by Doyle in Diseases of Swine, ed. by Dunne, 1964.

1960; Lussier, 1961); protruding ribs (Lussier, 1961); dark diarrhea, bloody stools, and temperatures ranging from 38.2 C. to 49.1 C. (Hofferd, 1936); and death. Though the disease appears in swine of all ages, Lussier (1961) found that 73% of the cases he observed occurred at 7 to 12 weeks of age. In an individual pig the course of the disease is usually 3 to 13 days (Doyle, 1939). Mortality averages about 40% in a herd (Birrell, 1957) but varies from 12% (Doyle, 1945) to 90% (Hofferd, 1936). Gastrointestinal diseases are rather common in swine, and these signs may be modified by nutrition, other pathogens, and husbandry practices. The cause of death is assumed to be due to dehydration and electrolyte loss.

Etiology. The etiology of swine dysentery has not been completely established (Birrell, 1957; Deas, 1960; and Davis, 1961b). However, it is generally accepted that Vibrio coli is probably the primary pathogen (Doyle, 1945 and 1948; James, 1947; Roberts, 1956a and 1956b; and Lussier, 1961). This organism can be consistently isolated from afflicted pigs; however, pure cultures fail to reproduce the disease (McNutt, 1947; James, 1947).

Pathology. Swine dysentery affects primarily the cecum and colon (Lussier, 1961). Other organs occasionally involved include the fundus of the stomach (Roberts, 1956a), the rectum (Lussier, 1961), and the mesenteric lymph nodes (Birrell, 1957). Birrell (1957) and Hofferd (1936) also reported excess fluid in the pericardial, pleural, and abdominal cavities. Histologically, the lesions of the gastrointestinal tract were found to range from catarrhal to hemorrhagic to necrotic, or a mixture of these, depending upon degree of inflammation and time of duration

(Lussier, 1961; Hofferd, 1936; Doyle, 1945; Roberts, 1956a; Deas, 1960; and Langham et al., 1952).

Therapy. The treatment of swine dysentery has revolved around 2 measures, antimicrobials and nutrition. Antimicrobials, including sulfamethazine (Boley, 1951), sulfathalidine (Boley, 1951; Graham et al., 1945), tylosin (Curtis, 1962; Miyat, 1964a and 1964b), hibitane (Lussier, 1961), bacitracin (Boley, 1951; Lussier, 1961), aureomycin (Salisbury, 1951), streptomycin (Salisbury, 1951; Birrell, 1957; and Doyle, 1954), nitrofurans (Roe, 1958; Davis,1961c; and Lussier, 1961), acetylarsen and stovarsol (Robinson, 1951), and sodium arsanilate (Boley, 1951) have all been used, though with few consistently beneficial results.

Nutritional measures which have been recommended usually suggest removal of corn and cereals and replacement with easily digested foods, such as buttermilk or sourmilk (Doyle, 1945; Hofferd, 1936; Aitken, 1954). Replacement therapy involving primarily sodium and chloride has been attempted by supplementing the feed at the rate of one pound of salt (sodium chloride) in each bushel of feed (Aitken, 1954) or drenching with a solution of 1/2 ounce each of sodium chloride and sodium bicarbonate in a pint of water per 100 pounds of live weight (Steenerson, in Doyle, 1945). An old system, believed to be of considerable value, consists of dissolving 10 pounds of salt in 50 gallons of water, soaking oats in it, and feeding this mixture for 4 or 5 days (Doyle, 1945).

Pathologic Physiology as Related to Electrolytes in Diarrhea of Man

Research reports on the influence of a specific colitis on electrolyte balance in the pig were not found in the veterinary literature. However, in the medical literature rather extensive studies have been summarized and may be used as a source of basic information applicable to this research undertaking. Stools from normal humans of all ages contain approximately twice as much potassium as sodium (Shohl, 1939). By introducing urine into colostomy pouches, Annis and Alexander (1952) reported that the colon could absorb sodium but not potassium.

The pertinent literature on electrolyte metabolism in man may be divided into references on infantile diarrhea and Asiatic cholera.

Infantile diarrhea. One of the first responses to an enteritis, regardless of the etiology, is the decreased transit time of ingesta through the gastrointestinal tract. This rapid passage results in decreased reabsorption of sodium; for example, Darrow (1946), the late and noted pediatrician, found that when the stool of babies became watery, the sodium loss always exceeded the potassium loss. Since sodium comprises over 90% of the extracellular base in the body (Black, 1953), it is apparent that a diarrhea of even short duration will seriously deplete sodium stores (Butler et al., 1933). This is especially true in infants, who have a greater percentage of their body weight in the highly labile extracellular space (Yannet and Darrow, 1938; Darrow, 1945). This decrease in sodium concentration will lead to a proportionate loss of water and, consequently, extracellular fluid volume (Harrison and Darrow, 1938; Darrow and Yannet, 1936). Such a loss of extracellular body water disturbs renal function and the osmotic equilibrium of the

extracellular and intracellular spaces and causes a shift of water from the intracellular to the extracellular compartment (Darrow, 1945; Elkinton et al., 1942; and Butler et al., 1933). The resulting cellular dehydration leads to potassium migration from the cell (Elkinton et al., 1948) and a rise in the serum potassium concentration (Darrow, 1946).

Darrow (1945) noted that loss of 30% to 50% of the extracellular electrolyte will cause shock and cellular anoxia (Darrow, 1946). This anoxia will also allow potassium to migrate from the cell and increase in the serum to the point of causing cardiac damage (Govan and Weiseth, 1946).

However, loss of cellular potassium is not due simply to cellular destruction and subsequent liberation of potassium, because the potassium to nitrogen ratio in the extracellular fluid is greater than the ratio of these constituents in the normal cell (Butler et al., 1933; and Elkinton et al., 1942).

When the cellular potassium concentration falls below an as yet undetermined point, sodium and hydrogen ions (Pitts, 1963) emigrate from the serum to the cell (Butler et al., 1933). With this replacement phenomenon, the cellular potassium concentration can drop to approximately half of normal (Darrow, 1956; Elkinton and Winkler, 1944). Though there is an approximately inverse correlation in the osmotic activity of these cations, there is no quantitative relationship between the net exchange of cell sodium and potassium and any internal alterations in osmotically active cell base. This is thought to be due to either osmotic deactivation or reactivation within the cell (Elkinton et al., 1948).

Another factor resulting from a decrease in the serum sodium concentration is a metabolic acidosis (Smith and Etteldorf, 1961), which also stimulates the movement of potassium from the cell to the serum (Darrow, 1948; Darrow, 1956; and Smith and Etteldorf, 1961). However, the poor ability of the kidney to retain potassium results in large quantities being lost in the urine (Darrow, 1945; Elkinton and Toffel, 1942). Darrow (1948) suggested that the acidosis is partially due to the fact that some bicarbonate from the serum follows the sodium intracellularly to form sodium bicarbonate. Thus, a cellular alkalosis is added to an already existing extracellular acidosis. When the serum bicarbonate drops, renal mechanisms increase the serum chloride concentration in order to maintain the electrical balance on that side of the membrane, a fact which also accentuates the extracellular acidosis (Pitts, 1963).

Therefore, a cycle is established: a loss of extracellular sodium results in an acidosis and a decrease in extracellular volume. These changes cause a decrease in cellular volume and a loss of cellular potassium, resulting in migration of sodium intracellularly to create an even more severe intracellular alkalosis. The result of this cycle -- depletion of cellular potassium -- will doom the organism unless proper therapy is initiated.

However, there are many factors, such as low potassium diets, injections of adrenal corticosteroids (Darrow and Miller, 1942), tissue breakdown, and mercurial diuretics (Blumgart et al., 1934), which lower the cellular potassium concentrations but do not result in the death of the organism. The movement of electrolytes in these examples is somewhat similar to the situation of a diarrhea in that sodium and

hydrogen ions still migrate intracellularly to replace the lost potassium. There are, however, several important differences: first, there is no massive depletion of serum sodium; secondly, the loss of cellular potassium is not initiated by acidosis, anoxia, or cellular dehydration; and thirdly, the effect of the intracellular migration of hydrogen ions causes the kidney to retain bicarbonate and excrete chloride (Pitts, 1963). This accumulation of bicarbonate causes an alkalosis which favors the replacement of cellular potassium at the expense of cellular sodium. Thus, the retention of renal function is the main difference affecting the outcome of the 2 conditions.

Although phosphorus levels in the stools have been reported to be 10 times the serum levels (Butler et al., 1933) and the muscles from babies in fatal cases of diarrhea indicate a loss of 10% to 20% of intracellular phosphorus (Darrow, 1946), Darrow stated that he could find no evidence of phosphorus loss in his metabolic studies (1946).

Loss of magnesium is a problem only in gastrointestinal disorders of many weeks' duration (Barnes et al., 1960), even though the physiology of this element parallels that of potassium (Wacker and Vallee, 1958b).

The only report located referring to serum calcium changes in diarrheas concerned calves. The conclusion was that losses are variable, but generally the values do not fall below low-normal (McSherry and Grinyer, 1954). However, in humans Rapoport (1947) found an alkalotic tetany associated with a hypocalcemia after the acidosis of diarrhea had been corrected with the use of parenteral fluids.

Asiatic cholera. Watten et al. (1959) reported slightly elevated sodium, chloride, and potassium values but lowered bicarbonate concentration in the serum of cholera patients regardless of the amount of fecal material excreted daily.

Therapeutics of Diarrhea in Man

Infantile diarrhea. Smith and Etteldorf (1961) evaluated several therapeutic regimens for infantile diarrhea and concluded that the regimen proposed by Govan and Darrow (1946) was best, with certain modifications. Briefly, the procedure, as modified by Smith and Etteldorf (1961), has the following basic steps: (1) combat shock with whole blood or plasma and an equal amount of saline. The object here is to overcome oliguria; (2) properly hydrate the baby and combat acidosis by giving 1/12M sodium lactate in 5% glucose; (3) repair the losses of electrolytes by administration of a solution of sodium chloride, potassium chloride, potassium lactate, and 5% glucose on a body weight basis. The solution supplies 122 mEq. sodium, 35 mEq. potassium, 104 mEq. chloride, 53 mEq. lactate, and 50 Gm. carbohydrate, totalling 314 mOsm. per liter.

In 1949, Darrow observed that the addition of potassium chloride to the replacement solution helped to restore cellular composition more quickly than without it. Since then, several investigators have reported that with full repletion of potassium, but no chloride, there was no change in the blood pH or bicarbonate concentration. However, with the administration and retention of chloride there was a prompt return of the blood pH and bicarbonate concentrations to normal and an increase in net alkali excretion (Aber et al., 1962; Atkins and Schwartz, 1962; and Kassirer, 1965).

Asiatic cholera. Phillips (1964), Director of the SEATO Cholera Research Institute, concluded from his studies that oral replacement therapy of water, potassium, and some of the bicarbonate losses was efficacious in treatment. Only sodium and chloride need to be given parenterally.

Summary

Reports from swine producing areas throughout the world indicate that swine dysentery is an important disease. Limited studies reporting on experimental cases of the disease have focused on evaluation and use of antibiotics.

To date there have been no reports examining electrolyte and water metabolism in swine dysentery. However, the pathologic physiology of infantile diarrhea and Asiatic cholera has been elucidated by several investigators, especially Darrow and Phillips. Both of these diseases have become more amenable to treatment since fluid and electrolyte losses have been sequentially and quantitatively characterized.

MATERIALS AND METHODS

General Experimental Design

A total of 18 healthy pigs was used in this project. Two pigs were used to establish the infection under conditions of the proposed research. Sixteen pigs were then used with the following experimental design:

1. Four pigs were to remain unexposed and serve as controls.
2. Four pigs were to be exposed and electrocuted at the time of the most severe diarrhea.
3. Four pigs were to die of the disease.
4. Four pigs were to recover.

Animals

For this study, 8 Yorkshire pigs from each of 2 healthy litters from the Michigan State University swine farm were used. All had been injected intramuscularly with 150 mg. of iron as iron-dextran at 3 days of age. They were weaned at 4 weeks of age.

Housing and Care

Three days post-weaning, the pigs were placed in individual, grated-bottom, galvanized cages containing separate crock containers for food and water. Two pigs from each litter served as controls and were housed in a separate room, though environmental conditions were similar. The ambient temperature in each room was 16 C.

The Michigan State University starter ration, minus antibiotics, was fed ad libitum to the pigs throughout the study. This is basically a corn-soybean meal ration with limestone, vitamins, trace mineralized salt, and vitamins added to provide the nutrient levels recommended by the National Research Council (1964). The average mineral concentrations were: sodium 0.27%, potassium 0.62%, phosphorus 0.54%, magnesium 0.13%, and calcium 0.71%. Only distilled water was available for drinking. Each pig was observed at 5- to 6-hour intervals. A daily record of water and food consumption, clinical condition, and behavior was maintained. An excrement-collection pan beneath each cage was covered with sheet vinyl in order to collect feces and urine. All pigs were weighed and rectal temperatures recorded at intervals throughout the study.

Infective Agent

The spiral colon of a pig experimentally infected with dysentery was supplied by Dr. Robert Berkman of Eli Lilly and Company, Greenfield, Indiana. A saline suspension of the colon contents and mucosal scrapings was used to expose two 3-week-old pigs in order to procure and maintain the Vibrio organisms until the test group was readied for inoculation.

Of these 2 pigs, the male developed a diarrhea within 3 days after exposure. Eight days following exposure his stool had become bloody. On this day the male was electrocuted, necropsied, and an inoculum suspension was produced for the test group by scraping the mucosa and contents of the spiral colon from this pig directly into 200 ml. of saline. Each of the 12 experimental pigs was given 10 ml. of the suspension as a drench and 10 ml. on the top of a small quantity of feed. All of the pigs were

fed half rations for the previous 24 hours; therefore, the pigs in the test group consumed their contaminated feed within 20 minutes.

Within 10 days following initial exposure, only one pig had sickened and died; the remainder were clinically healthy. At this point there was concern that the healthy pigs would not contract the disease; therefore, they were inoculated with the contents of another spiral colon supplied by Eli Lilly and Company according to the procedure previously described. No other bacterial pathogens were found on *in vitro* cultures of either inoculating suspension. The *Vibrio coli* organisms could be seen on direct microscopic examination of the inoculum, but were successfully cultured on only about 50% of the attempts.

Blood and Serum Analyses

Eight milliliter samples of blood were collected from the anterior vena cava by the method of Carle and Dewhirst (1942). All pigs were bled 2 days prior to exposure and 2 and 7 days postexposure. An attempt was made to collect samples from each pig at the initiation of the bloody diarrhea, 2 to 3 days after its onset and when the animal was prostrate.

Serum for electrolyte determinations was obtained by placing 6 ml. of blood in an acid-washed centrifuge tube and allowing it to clot. The serum was separated by centrifugation at 2500 *g.*, transferred to an acid-washed vial, and stored at 4 C. until analyzed.

Serum sodium and potassium were determined by flame emission spectrophotometry, using a Jarrell-Ash Model 82-516 with a Hetco (total consumption) burner and an air-hydrogen flame. A serum sample of 0.1 ml. was diluted to 10 ml. with deionized water. Both sodium and potassium

analyses were made from the same dilution of serum. Sodium was determined at a wave length of 5899.9 angstroms and potassium at 7664.9 angstroms.

Serum chloride values were determined by the following micro-modification of the colorimetric method described by Schales and Schales (1941): to 0.1 ml. of serum one drop of s-diphenylcarbozone and one drop of 2N nitric acid were added. Sufficient mercuric nitrate was added to the unknown solution to obtain a permanent blue-violet color.

Blood from the sample drawn was allowed to flow into heparinized capillary tubes and immediately analyzed for partial pressure of CO_2 (pCO_2) with a Radiometer pHM27* and the pCO_2 Electrode, type E 5036. Blood bicarbonate concentration was then determined by utilizing the Sigaard-Andersen alignment nomogram.

Inorganic phosphorus was determined by the application of the spectrophotometric method of Gomori (1942).

Serum calcium and magnesium were determined by atomic absorption spectrophotometry, using the Jarrell-Ash instrument. Calcium was determined on a trichloracetic acid supernatant fluid after a dilution with a strontium chloride solution in order to provide a final strontium concentration of 10,000 p.p.m. to overcome phosphate interference. Calcium absorption was measured at 4226.7 angstroms. For magnesium a 0.1 ml. sample of serum was diluted with 9.9 ml. of deionized water. The absorption was measured at 2852.1 angstroms.

Immediately after the blood was collected the pH was determined with the pHM27 and a Micro-Electrode Unit, type E 5021. Hemoglobin

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*Radiometer A/s, 72 Emdrupvej, Copenhagen NV, Denmark.

estimation was by the cyanmethemoglobin method (Benjamin, 1964). Hematocrit was determined by the micro-hematocrit method (McGovern et al., 1955). Total and differential white-cell counts were determined as described by Benjamin (1964).

Blood volume was determined according to the method described by Ramirez et al. (1963) at 4 days prior to exposure, 3 days postexposure, and when the pig was prostrate.

Electrolytes in Excreta

Samples composed mainly of fecal material were collected over a 24-hour period at the initiation of the project and again at the time of most severe dysentery. For analysis a 2 Gm. aliquot (wet basis) was digested in 50 ml. of concentrated nitric acid (HNO_3) with heat until a residue of less than 1 ml. remained. This residue was digested further with 7 ml. perchloric acid ($HClO_4$) and heat; the volume was then reduced to approximately 1 ml. by evaporation. Sufficient deionized water was added to the residue to bring the weight of the sample to 100 Gm. From this diluted sample, electrolyte concentrations were determined incorporating the same equipment and similar techniques as previously described for serum electrolyte concentrations. However, calcium and magnesium analyses were conducted on samples to which sufficient strontium was added to provide a final concentration of 8000 p.p.m.

Histopathologic Technique

At necropsy, samples of lung, thyroid, heart, spleen, liver, kidney, pancreas, adrenal, fundic portion of the stomach, duodenum, ileum, cecum, spiral colon, and rectum of each pig were fixed in 10% formalin and prepared for examination according to methods described in the Manual

of Histologic and Special Staining Technics of the Armed Forces Institute of Pathology, Washington, D.C. (1957).

RESULTS

The research generally proceeded as planned. The control pigs remained clinically healthy throughout the experiment and gained at an average rate of 0.22 kg. per day. However, the original experimental design for the exposed pigs had to be modified for 2 reasons: (1) none of the pigs infected with swine dysentery recovered, and (2) the clinical signs became apparent at such variable hours that experienced help was not always available to assist in the planned collection of blood samples. For these reasons the analysis was carried out with the Fisher t test of significance between 2 sample means. Values of the normal period and those of the prostrate period were collated for each of the infected animals, while first and final values were compared for the control pigs.

Clinical Signs

Incubation time for the disease ranged from 6 to 23 days, with an average of 13 days. Generally, the first indication that infection had occurred was a rough hair coat. This was followed almost immediately by an arched back, gaunt abdomen, and a reluctance to move (Figure 1). There was no sign of mental depression at this time. Temperatures ranged from 38.5 C. to 41.0 C. and averaged approximately 40.0 C., which was about 2.0 C. higher than the controls.

The rough hair coat usually preceded the onset of the bloody diarrhea by 1 to 2 days. The character of the feces usually progressed from a

Figure 1. Uninfected pig (background) contrasted to an infected pig that has a rough hair coat, gaunt abdomen, and arched back.

normal, well-formed type to a soft specimen and finally to a bloody mucoid sample. This generally occurred within a period of 12 to 16 hours. Due to the rapid onset of the dysentery, there was frequently no evidence of the intermediate stage in this progression. As a rule, once the melena began, the pigs remained bright and alert for about 2 days; however, food consumption dropped to approximately 60% of normal (Table 1).

Two to three days after the melena began, other signs were observed: (1) temperature returned to normal (38.9 C.); (2) food consumption was reduced; and (3) mental depression was apparent. By the 4th day of the bloody diarrhea, food consumption had usually ceased.

Water consumption increased by about 25% 2 days before any signs of sickness were evident, returned to normal as the diarrhea began, and then increased by 25% again 2 days following the start of the melena. After the 2nd increase, water consumption subsided to almost nothing within a day or two. Once the pigs had ceased drinking, they began to exhibit cyanosis of the ear tips, abdomen, and inguinal region. A short period of posterior ataxia preceded prostration. At the time of prostration the average temperature was 36.5 C., and the anal sphincters were relaxed.

The exposed pigs lost an average of 0.32 kg. of weight per day of diarrhea, which resulted in a loss in body weight of 32% from the onset of diarrhea to death. The course of the disease in the average pig was 4.5 days.

Blood Values

Information on the serum electrolytes, blood pH, hemoglobin, and hematocrit is summarized in Table 2. A more detailed explanation of the results for these items follows.

Table 1. Average food and water consumption for the 48-hour period before dysentery to the 48-hour period after the onset of dysentery.

	Pre-dysentery	Dysentery
Food (Gm.)	550	330
H_2O (ml.)	9400	1130

Table 2. Serum electrolyte and hematologic data in experimental vibrionic colitis.

	Sodium (mEq./L.)	Potassium (mEq./L.)	Bicarbonate (mEq./L.)	Chloride (mEq./L.)	Phosphorus (mEq./L.)	Magnesium (mEq./L.)	Calcium (mEq./L.)	Blood pH	Hemoglobin (Gm./100 ml.)	Hematocrit (vol.%)
Experimental (normal)	146.5 (24)*	5.10 (24)	29.1 (24)	104.2 (24)	3.56 (24)	3.11 (24)	5.51 (24)	7.33 (24)	11.29 (24)	38.1 (24)
Control (normal)	153.4 (8)	5.48 (8)	30.6 (8)	102.3 (8)	3.91 (8)	2.36 (8)	5.97 (8)	7.24 (8)	12.07 (8)	40.9 (8)
Experimental										
Day 1	146.6 (2)	5.92 (3)	47.7 (3)	103.6 (3)	4.91 (3)	2.45 (3)	5.42 (3)	7.29 (3)	8.40 (3)	27.7 (2)
Day 2	149.9 (3)	7.10 (3)	29.6 (3)	102.1 (3)	5.66 (3)	3.26 (3)	5.71 (3)	7.21 (3)	10.33 (3)	38.0 (1)
Day 3	132.2 (3)	6.75 (3)	20.8 (3)	94.4 (2)	6.78 (3)	3.18 (3)	6.00 (3)	7.16 (3)	9.53 (3)	31.0 (2)
Day 4	120.0 (6)	9.67 (6)	14.8 (6)	93.8 (6)	10.51 (6)	3.83 (6)	6.06 (5)	7.10 (6)	11.10 (3)	36.0 (4)
Day 5	113.8 (3)	8.86 (3)	14.4 (3)	82.9 (3)	9.49 (3)	3.83 (3)	6.93 (3)	7.22 (3)	10.90 (2)	37.1 (3)
Day 6	114.6 (1)	10.74 (1)	10.7 (1)	85.3 (1)	8.93 (1)	4.50 (1)	5.15 (1)	7.15 (1)	7.00 (1)	26.0 (1)
Day 7	128.8 (1)	7.54 (1)	22.6 (1)	85.7 (1)	7.31 (1)	3.62 (1)	5.25 (1)	7.35 (1)	9.20 (1)	32.0 (1)
Experimental (prostrate final)	117.5 (9)	10.41 (8)	14.6 (8)	81.9 (8)	10.76 (8)	4.23 (8)	6.22 (8)	6.91 (8)	9.50 (5)	33.8 (8)

Table 2--continued

	Sodium (mEq./L.)	Potassium (mEq./L.)	Bicarbonate (mEq./L.)	Chloride (mEq./L.)	Phosphorus (mEq./L.)	Magnesium (mEq./L.)	Calcium (mEq./L.)	Blood pH	Hemoglobin (Gm./100 ml.)	Hematocrit (vol.%)
Control (final)	152.5 (3)	7.13 (3)	35.0 (4)	100.0 (3)	6.47 (2)	3.56 (3)	6.30 (2)	7.06 (4)	11.15 (4)	37.2 (4)
Statistical significance**	P>0.2, 0.3	P>0.2, 0.3	P>0.05, 0.1	P>0.2, 0.3	P>0.1, 0.2	P>0.9	P>0.7, 0.8	---	P>0.05, 0.1	P>0.1, 0.2

* ( ) = number of samples utilized to determine the average.

** Comparison of the control versus prostrate values.

Serum electrolytes. Sodium values began to decline on the 3rd day of dysentery and continued to decline until death, at which time the average concentration was 117.5 mEq./L. The average value of the control pigs at the termination of the project was 152.5 mEq./L. Potassium concentration rose slowly from 5.1 mEq./L, the approximate normal value, to 10.4 mEq./L, the average value at prostration. This was almost 3.3 mEq./L above the value of the control pigs at termination. Chloride levels remained near the normal average of 104 mEq./L until the 3rd day of the dysentery, when they declined to an average of 81.9 mEq./L at prostration. The controls maintained a rather consistent value throughout the research trial.

Initially the serum bicarbonate values in the exposed pigs averaged approximately 30 mEq./L. At prostration the average concentration in the infected pigs was approximately 14.6 mEq./L. This was well below the 35.0 mEq./L average for the control pigs. Serum phosphorus increased slowly from the initial values of 3.56 mEq./L to a concentration of about 8 mEq./L during the dysentery and then increased rapidly during prostration to an average of 10.6 mEq./L. Magnesium and calcium values in the infected pigs did not vary noticeably from values of the control pigs.

Blood pH. An average of the beginning blood pH values was 7.33 but declined from the 2nd day of the dysentery until prostration. At prostration the average value was 6.91.

Hemoglobin and hematocrit. Hemoglobin and hematocrit decreased from 11.7 Gm./100 ml. and 37.7% to 9.5 Gm./100 ml. and 33.8%, respectively. The final values in the control pigs were 11.2 Gm./100 ml. for hemoglobin and 37.2% for hematocrit.

Total white blood cell (WBC) and differential. There was no noticeable difference at any time during the research in the total WBC between the controls and the infected pigs. However, in the prostration samples, a large number of immature neutrophils was noted.

Blood volume. The initial values for blood volume averaged 10.2 ml./100 Gm. of body weight for both groups. At project termination, values for the uninfected pigs had risen to 12.2 ml./100 Gm. of body weight, while the infected pigs at prostration averaged 9.9 ml./100 Gm. of body weight.

Electrolytes in Excreta

As the relative amount of sodium increased in the dysentery feces, phosphorus and magnesium either remained the same or decreased. There was no statistical significance in the ratios of the other electrolytes to each other.

Pathology

Gross. The pigs which died of swine dysentery had several external signs in common -- cyanosis, diarrhea, and emaciation.

It was noted upon opening the pleural and peritoneal cavities that the lesions primarily involved the gastrointestinal system. The most outstanding consistent lesion was a severe hemorrhagic colitis (Figure 2). Frequently the congestion and hemorrhage were so severe that a dark purple color could be observed from the serosal surface. However, one pig had only a catarrhal colitis. In addition to the colitis, 6 pigs had gastritis. One pig had a severe case, while 5 others had some hyperemic foci in the fundic portion of the stomach. Pneumonia with a

Figure 2. Ileo-ceco-colic junction of infected pig. Fibrinonecrotic membrane in colon (a); hyperemic cecum (b); and normal ileum (c). The mucosal surface had been washed free of hemorrhagic debris.

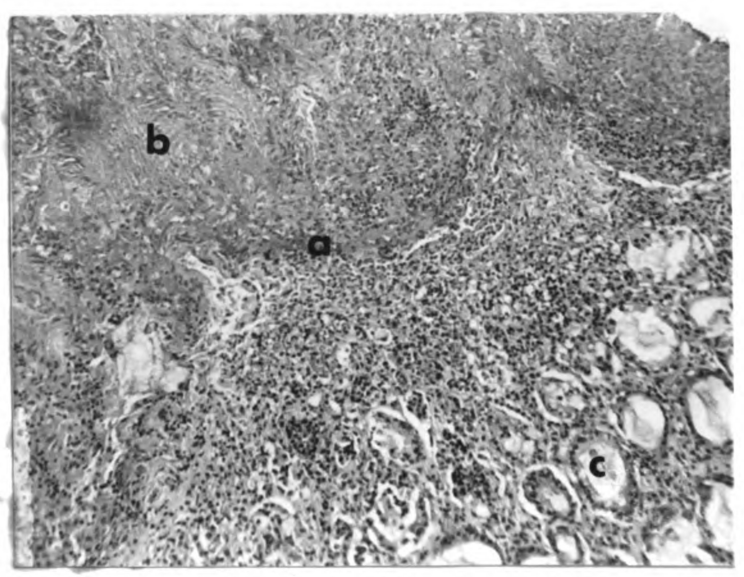
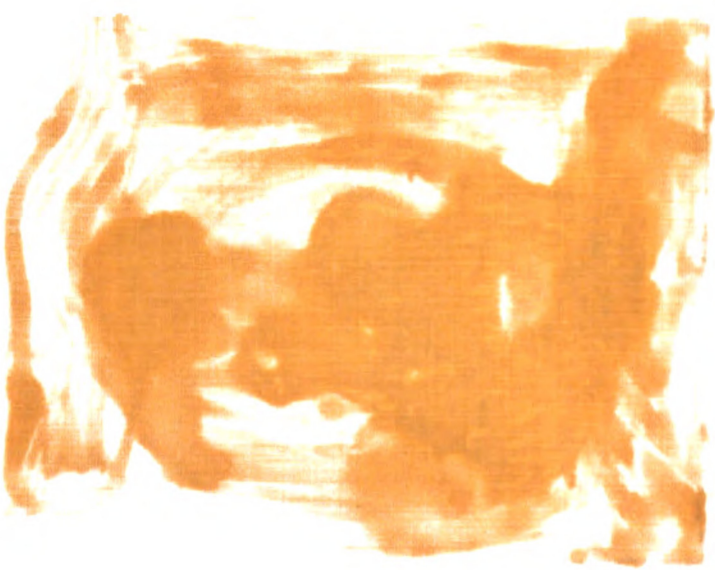


Figure 3. Colon of infected pig. Necrosis and denudation of surface epithelium (a); fibrinonecrotic membrane (b); and mucinous degeneration of the mucosal glands (c). Hematoxylin and eosin. x 175.



fibrinous pleuritis was observed in one pig. In several pigs the heart appeared rounded and flaccid; however, no gross anatomical defects were found. Occasionally the liver exuded considerable blood from the cut surface.

Histologic. The mucosal surface of the colon and cecum was usually necrotic and hemorrhagic. In some sections of the cecum there were neutrophils and/or lymphocytes near the zone of inflammation, but usually inflammatory cells were not seen. Though the inflammation of the colon wall was very severe, it rarely involved the deeper portions of the crypts or lamina propria. The lumen of the colon was commonly filled with necrotic debris, ingesta, mucus, and sloughed epithelial cells (Figure 3). The mechanism by which the blood cells entered the lumen of the bowel was not elucidated.

The remainder of the gastrointestinal tract was not as severely involved as the colon. In the fundic portion of the stomach there were several small areas which appeared to be extremely congested and denuded of epithelium; some of the congestion was due to venous thrombosis in the submucosa. However, few inflammatory cells were observed (Figure 3).

Occasionally the liver was congested, blood being apparent in the sinusoids. In some tissue sections it was difficult to detect the outline of the hepatic cells, though the nuclei appeared normal. In several kidneys there were tightly packed glomerular tufts; the nuclei were hyperchromatic; and there were few erythrocytes in the vessels. The convoluted tubule cells were usually swollen, frequently occluding the lumen. In the adrenal gland, pyknotic nuclei and foamy cytoplasm were in the zona reticularis.

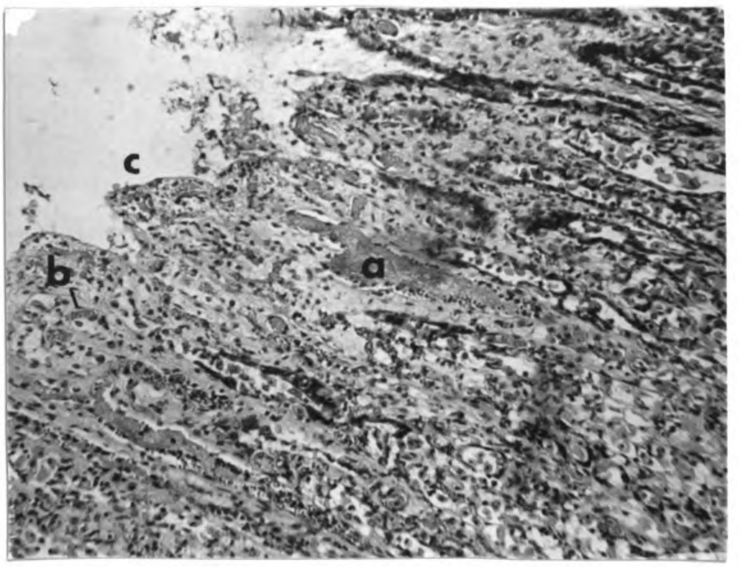


Figure 4. Stomach of infected pig. Thrombi (a) and congestion (b) in the vessels of the submucosa and loss of mucosal epithelium (c). Hematoxylin and eosin. x 175.

On histologic examination, there were no pathologic lesions observed in the following organs: thyroid, spleen, pancreas, lung, heart, mesenteric lymph nodes, and small intestine.

DISCUSSION

Clinical Signs

The clinical signs of experimentally produced swine dysentery were similar to those reported from field cases. However, in this research, the morbidity and mortality rates were higher than those reported by Roberts (1956a). Though there was an early febrile response, temperatures usually returned to normal by the 2nd day of the melena. It was at this time that the serum electrolyte changes became evident.

Serum Electrolytes

Serum electrolyte changes seemed to follow the initial decrease of serum sodium. This was especially apparent for chloride, which began to decrease rapidly on the 3rd day of dysentery. As sodium and chloride declined, the bicarbonate and pH also decreased. At the time of prostration, serum potassium and phosphorus values increased sharply, while at the same time pH and bicarbonate were at their lowest value. The blood volume was markedly decreased at prostration.

The ratio of sodium loss to both magnesium and phosphorus loss in the dysentery stools was significantly greater than in normal stools.

Acidosis probably resulted from the loss of extracellular sodium to the bowel contents and respiratory depression. It is possible that this acidosis, as well as a partial anoxia resulting from the decreased extracellular fluid volume, resulted in the emigration of potassium -- and probably phosphorus -- from the cell.

Though the white blood cell count did not vary markedly, there was a higher percentage of immature neutrophils in the diseased pigs.

Pathology

Gross. The gross pathology of swine dysentery in this research was quite similar to that reported from field cases. However, severe cyanosis of various portions of the body was noted even before the animals were prostrate. Frequently, the mucosa of the colon was so obviously inflamed and so heavily covered with fibrinous exudate that it was suggestive of a disease of swine commonly known as "necro".

Histologic. It seems unusual that a disease which kills young pigs does not have a more invasive course and greater tissue changes. Viewed grossly or histologically, the lesions are marked, but they primarily involve the superficial aspects of the colon.

Small foci in the fundic portion of the stomach were areas of coagulation necrosis, probably due to the venous infarction occurring in the vessels of the submucosa. The etiology and pathogenesis of these lesions is unclear.

The adrenal gland contained pyknotic nuclei in the cells of the zona glomerulosa and a few in the cells of the zona fasciculata. The cytoplasm of these cells appeared to be foamy, but the outline was distinct.

This research indicates that a specific infectious colitis can be experimentally produced in swine and might serve as a useful research model for additional work as applied to comparative medicine.

SUMMARY

A total of 16, 5-week-old Yorkshire pigs was used to study the pathology and changes in serum electrolyte and hemogram values due to an infectious colitis caused by Vibrio coli.

The clinical signs and gross and histologic lesions experimentally produced were typical of those noted for naturally occurring field cases. The clinical signs consisted of dehydration, anorexia, cyanosis, dysentery, prostration, and death. The lesions were primarily a necrotic-hemorrhagic colitis and typhlitis.

As the dysentery progressed, hematocrit and hemoglobin increased significantly, while blood pH and blood volume decreased. Serum sodium, chloride, and bicarbonate declined on the 3rd day of the dysentery; potassium and phosphorus increased at prostration. Calcium and magnesium did not vary markedly from the controls.

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