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Hog Cholera

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**AGRICULTURAL
EXPERIMENT STATION**

**SOUTH DAKOTA
STATE COLLEGE OF AGRICULTURE
AND MECHANIC ARTS**

VETERINARY DEPARTMENT

Hog Cholera

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Hog Cholera

E. L. Moore

T. B. Kelly

Hog Cholera is an infectious disease of swine characterized by a high fever and more or less extensive congestion and hemorrhages of the lymphatic glands, the intestines, kidneys, lungs, spleen, heart and liver. Various other names have been applied to this disease, such as pig typhoid, swine fever, pneumo-enteritis, blue disease, etc., but it is most frequently referred to as hog cholera.

The annual loss of pigs from this disease in the United States amounts to thousands of dollars and its ravages are a constant source of discouragement to breeders of pure-bred stock as well as those who make a special business of feeding hogs for the market. It is impossible to determine the extent of the disease in South Dakota; and while in general the disease is confined to the southeastern portion of the state, yet outbreaks have been reported in various other sections of the state, including the most northern counties.

Ever since the advent of this disease in the United States in 1833 investigations have been almost constantly conducted in an effort to determine the cause. Considerable work was done along this line by Law and Detmers in the seventies. In 1885 Salmon and Smith isolated and described an organism, *Bacillus cholerae suis* which was designated as the causative agent. *B. cholerae suis* from this time until 1903 was considered by the majority of investigators to be the specific cause. During this period certain discrepancies were shown to exist which stimulated repeated inquiries with reference to this disease. In 1903 de Schweinitz and Dorset showed that the disease known as hog cholera was due to an infecting material contained in the body fluids which was capable of passing through the finest porcelain filters. Later experiments carried on by Dorset, Bolton and McBryde showed that the virus found in the blood of hogs presenting typical lesions of cholera, when passed through an exceedingly fine filter, was still capable of producing the disease in its characteristic form when injected into susceptible hogs. This filtrate contained no organisms of *B. cholerae suis*, as was shown by cultural and microscopical tests. While uniform results in reproducing the disease and also in the production of immunity have been obtained with the germ free filtrate, such has not been the case

with the use of pure cultures of *B. cholera suis* alone. As a result of investigations along these lines it is now generally accepted that hog cholera is due to a filterable virus contained in the blood of affected animals. Just what role *B. cholera suis* plays in connection with this disease is not fully understood but it is believed to be more in the nature of a secondary invader.

Symptoms. The symptoms of hog cholera vary widely. The disease may take the acute or chronic form. Usually the first outbreaks are marked by a high virulence and death rate, the hogs dying off very rapidly. As a rule this period is followed by a decline in the strength of the virus and the animals linger several days or weeks before succumbing to the effects of the disease. The period of incubation of cholera, that is, the time which elapses between actual infection until noticeable symptoms appear, is from three or four to eight or ten days. In the acute form, the hog is usually very sick in eight days after infection and dies within four or five days thereafter. However, it is not uncommon for the animals to die very suddenly without manifesting any indications of being sick. The first symptoms are generally a loss of appetite, quite a pronounced fever, the hog appearing very dumpish. Most of the time the animal lies huddled up in the corner and only moves about when thirsty or when driven out of his resting place. He stands with his back arched, he may become lame in the hind legs, his snout gets very dry and the eyelids are often glued together with a sticky muco-purulent discharge. If the outbreak is marked by a high virulence, the ears, snout, belly and buttocks begin to flush and the animal dies within a few hours. These flushed areas may become so congested that they have a purplish appearance, from which undoubtedly came the common name "purples," used in some localities, for this disease. Diarrhoea may occur after the hogs have become thoroughly sick and may persist until the end. A bloody diarrhoea not infrequently occurs. In some cases, however, there may be constipation, the manure being firm, and covered with mucous.

Practically the only difference between the acute and chronic form of cholera is the duration of the disease and the lack of feverishness and flushing in the chronic type. With the chronic form, the hog becomes unthrifty and lingers perhaps for three or four weeks before dying. The coat becomes rough and the animal eats perhaps a little every day but as time goes on he gradually becomes thinner and weaker and finally dies of exhaustion.

The symptoms being varied and at best not especially characteristic, a post mortem examination must be resorted to in order to make an accurate diagnosis.

POST MORTEM APPEARANCES

External Appearances. In the acute type of cholera, flushing of the ears, snout, throat, belly and buttocks is most commonly found. Sometimes this flushing is confined to the belly and buttocks, perhaps the throat. Sometimes only the buttocks and throat are affected, or again, no flushing may be evident.

Lungs. In well defined cases of cholera there may be several collapsed areas which are noted by their dark color. These collapsed areas may be small or they may be quite extensive, but are not essentially characteristic of the disease.

The darkened portions are firm and are found in the upper part of the lungs. Often the lungs are badly congested owing to the escape of blood from the minute capillaries. When such is the case the lungs are covered with minute red spots (petechiae) about the size of the head of a pin, or there may be numerous purplish spots or blotches (ecchymoses) much larger in size.

Heart. The heart as a rule shows very few lesions. In some of the cases, however, there may be numerous petechiae

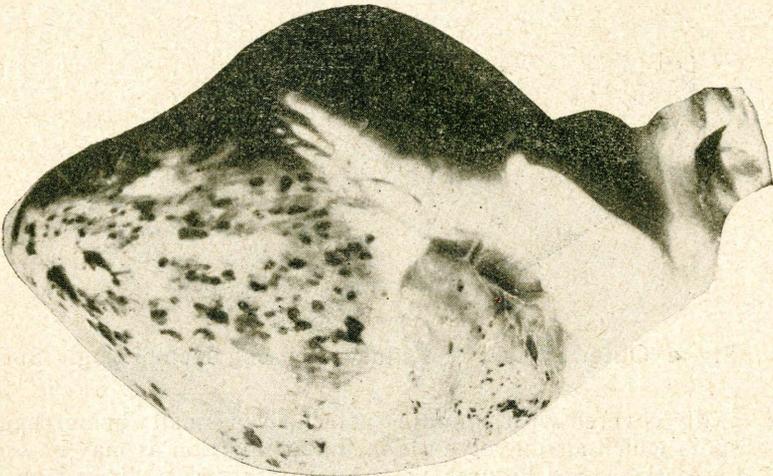


Fig. 1. Petechiae on the Heart of a hog Affected With Cholera

and hemorrhagic spots found on this organ. Figure 1 shows these lesions on the heart of a hog killed eight days after being

injected with a highly virulent strain of cholera producing serum. The lesions of this particular hog were excellent.

Spleen. Spleen or "milt" as it is commonly called, presents various lesions. Frequently no abnormal features appear. It may be more or less congested, often enlarged and sometimes very soft, so that slight pressure easily ruptures it. Hemorrhagic spots may be present. In chronic cases, the spleen may be much enlarged, even to several times its normal size, or it may be slightly smaller than normal, with no particular changes visible to the naked eye. In acute cases the spleen is soft, gorged with blood and often enlarged. It also may show hemorrhagic spots or areas underneath its surface. In chronic cases of the disease there may be an increase in the connective tissue of the spleen, in which case it will be firm and may be smaller than normal.

Stomach. This organ rarely shows any lesions except in cases where the hemorrhages are extremely severe. In that case, the inner or mucous coat is badly congested, while the outer membrane may or may not show hemorrhagic spots. In other case it may show an ulceration of the mucous coat.

Kidneys. The lesions of the kidneys vary considerably. Sometimes the cortex or outer dark red portion may be lit-

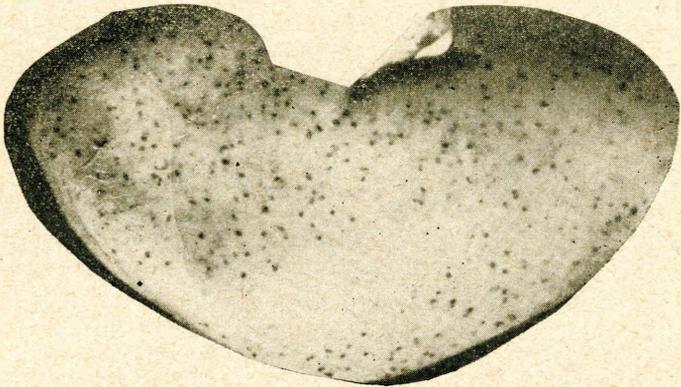


Fig. 2. Outer Surface of Kidney Showing Hemorrhagic Spots

erally covered with minute petechiae or small hemorrhagic spots which also involve the medullary portion as may be seen on section. (See Figures 2 and 3.) In other cases there may be only a few such petechiae on the cortex. Where hemorrhages are extremely severe, clotted blood is often found in the interior. It is not at all uncommon to find no particular lesions.

Bladder. The lesions of the bladder may vary from none at all to many petechiae on the mucosa or inner lining. Occasionally congestion and hemorrhage may be so severe that the walls of the bladder become much thickened and "knobby"

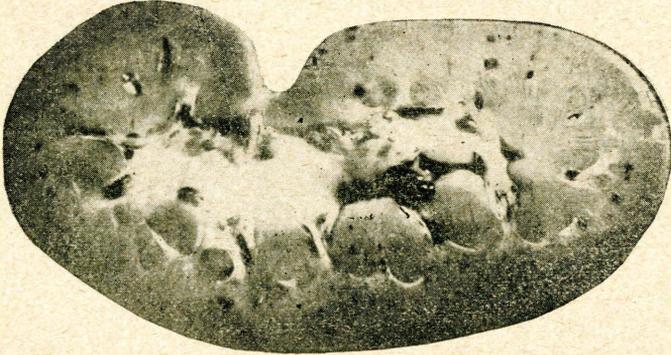


Fig. 3. Longitudinal Section of Kidney Showing Hemorrhagic Spots on Cortex and Medullary Portion

and dark purple in color. However, such instances have occurred only a few times in our work at this station where several hundred hogs have been killed for cholera, and the lesions if present consist of small hemorrhagic spots on the mucosa.

Intestines. In the acute form of cholera the mucous coat of the intestines, particularly in the region of the ileo-cecal valve, the place where the small intestine opens into the large intestine, may be more or less congested and covered with petechiae. In some hogs clotted blood may be found in the large intestines. The most constant and characteristic lesions of the intestines, however, consist in the presence of erosions or ulcers of the mucous membrane. These appear as somewhat circular button-shaped ulcers with a firm dirty-grey or greenish-yellow center standing out plainly from the surrounding mucous membrane. (See Figure 4). In some cases these necrotic changes become so extensive that large areas of the intestinal mucosa are involved.

Lymphatics. The lymphatic glands to which special attention is paid in hog cholera, are the inguinal glands, situated beneath the skin on the inside and high up on the thighs; the mesenteric glands, or those along the intestines; retroperitoneal and lumbar, or those lying near the back wall of the abdominal cavity; the bronchial and mediastinal, those lying in the region of the lungs and heart; and the lymphatics found in the neck near the angle of the jaw. The changes occurring in

the lymphatic glands consist of enlargement and congestion. These changes vary from showing bright red lines of congestion to cases in which the glands are almost black. If cut into it will be found that the congestion has occurred principally in the exterior of the gland.

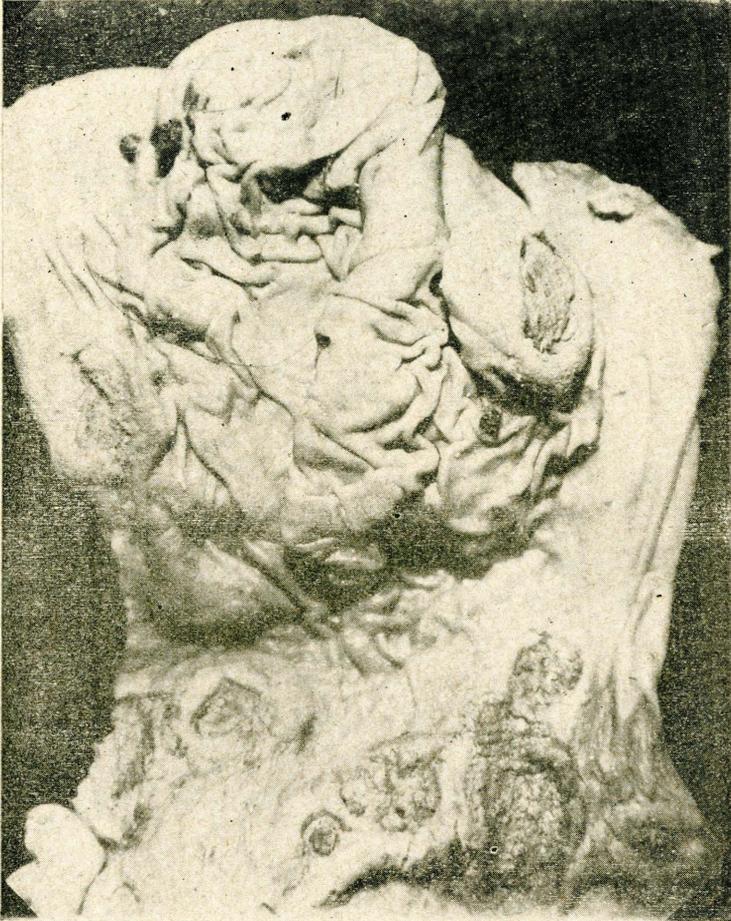


Fig. 4. A Portion of the Cecum Split Open Longitudinally So As to Expose the Ulcerated Mucous Membrane

These post mortem changes or lesions may not all be found in the one individual and for the most part a number

of animals must be examined before the picture as described is completed in its entirety.

PREVENTION OF CHOLERA BY ARTIFICIAL IMMUNITY

Cures and vaccines for hog cholera have in the past and are still flooding the market. Many widely advertised cures for cholera have proven to be of no value. When it was believed that *B. cholera suis* was the causal factor of the disease several attempts were made to produce a successful vaccine or antitoxin through the use of this organism. While a slight degree of success attended these efforts, nevertheless, they were not always satisfactory. A sufficient explanation of the failure of an antitoxin made with *B. cholera suis* was not forthcoming until it was proven conclusively that cholera resulted from a filterable virus. As already noted this filterable virus, which is to be found in the blood of animals sick with the disease, invariably produces typical cases of hog cholera, which was not true with *B. cholera suis*. As soon as the cause of cholera was rightfully determined, a substantial basis was given upon which to experiment in the production of a vaccine which would prevent the disease.

In almost any outbreak of cholera there is a small percentage of hogs that survive the disease. It is well known that in such cases the surviving animals possess an immunity to cholera which permits them to withstand an attack of a highly virulent strain of the disease. Nevertheless, the blood of these hogs possesses little protective properties when injected into susceptible hogs, as they will contract the disease when they are afterwards exposed to cholera. The problem to be solved then was to increase the protective properties of the blood of immune hogs. This is done by injecting varying amounts of virulent blood, that is, blood which is capable of producing cholera, directly into the blood system of the immune. In the first experiments only small doses were given, but as progress was made the dose of virulent blood necessary to produce a potent serum which would protect a non-immune was considerably increased. Much careful experimenting has been done along this line and has resulted in a standard method of producing a protective serum, the credit of developing which belongs to Dorset, Niles and McBryde.

METHOD OF PRODUCING POTENT SERUM

The method of producing a protective serum for hog cholera consists in hyperimmunizing immune hogs, either

natural or artificial immunes, with proper amounts of virulent or disease-producing blood. Virulent blood is obtained from small hogs which have been inoculated with a small amount of cholera virus, usually 2cc. The pigs used for the production of virulent blood weigh from forty to fifty pounds and are injected in the thigh with the virus. If the virus is strong enough the injected animals sicken in the usual length of time, that is eight days, and should die in from ten to fifteen days. Just before the animals succumb to the disease they are killed and the blood collected. The whole process of collecting and handling of blood is conducted with as strict aseptic precautions as possible. After the blood has been drawn it is allowed to clot. The serum is then separated from the clot and is ready to use for hyperimmunization.

The hogs used for hyperimmunizing weigh from 125 to 200 pounds. They are artificially immunized either by passing successfully through the disease or by laboratory methods. The animals are securely fastened and are then injected either intravenously (directly into the ear vein), intraperitoneally (in the abdominal cavity) or subcutaneously (just beneath the skin). In order to produce a fully hyperimmunized hog, the intravenous method requires 5cc. of virulent blood for every pound of body weight, while the other two methods require 10cc. The virulent blood used for this purpose must be standardized, that is, it must kill a small hog within fifteen days. A virus which does not possess this degree of virulence is not employed in making potent serum. Supposing a hog weighed 150 pounds he would require 750cc. of virulent blood intravenously or 1500cc. intraperitoneally or subcutaneously to be completely hyperimmunized. The intravenous method is the most desirable since it requires less serum, and any undesirable after effects, such as abscess formations, are avoided. In addition to this, our experience indicates that a more uniformly potent serum can be obtained by the first method. Its chief objection lies in the difficulty of inserting a needle directly into one of the ear veins. This may be largely overcome by wrapping some cotton wrung out of hot water directly around the base of the ear, maintaining it in position by slipping a rubber band over the cotton. The heat and the constriction of the rubber band dilate the ear veins so that the needle can be readily inserted. Both cotton and band are removed during the injection of the blood.

When the first experiments were conducted in making potent serum two methods of injecting the virus were employed, i. e., the slow and quick methods. The only difference between

the two is in the amount of each dose of virulent blood and the time elapsing between ensuing injections. A hog hyperimmunized by the quick method should receive the required amount at one injection. With the slow method the hog receives only a very small amount of virulent blood at the first dose and after a week or so receives a somewhat larger dose. This in turn is followed by a still larger dose after several days and so on until the animal has received enough to be completely hyperimmunized. The slow method is of no advantage and has been entirely supplanted by the quick method. It will therefore be seen that by the method of hyperimmunization we have largely increased the protective substances contained in the blood of the hog so that its serum is potent, that is, it will protect a susceptible hog from cholera if injected in proper amounts.

After a week or ten days the hyperimmunes are bled from the tail to secure the potent serum. A small piece of the tail is clipped off and the blood collected in a sterile dish, the animal being bled as much as he can stand without distress. The clot is removed from the serum to which is added a sufficient quantity of a 5 per cent solution of carbolic acid to make the serum contain 0.5 per cent carbolic acid. It is then stored in sterile bottles and kept in a dark cool place and should retain its efficiency for several months without deterioration. A week after the first bleeding, the second bleeding is made in the same manner as the first. Three or four bleedings of each hyperimmune are made at weekly intervals and the animal is then killed and all his blood collected or else it is rehyperimmunized. The amount of virulent blood required for rehyperimmunization is just half of that used the first time. The potent serum is then collected exactly as in the first case. This method of rehyperimmunizing and bleeding may be continued as long as any tail is available.

TESTING POTENT SERUM

The next step in the manufacture of potent serum is the testing of the potency. This is done by taking the minimum dose necessary to protect a hog in the field and injecting it into a small pig. The usual method is to take three hogs weighing about fifty pounds, preferably from the same litter if possible, giving each of them 2cc. of virulent blood on the inside of the right thigh and then injecting two of the pigs in the left thigh with 15cc of potent serum. Thus we have one hog that has received only virulent blood while the other two have received in addition to the virulent blood the potent serum, which should protect them from cholera. The animals are

then placed in a clean, disinfected pen and observed through a period of about three weeks. The virulent hog, that is, the hog which has been given the virus alone, should be ready to kill within fifteen days, while the others remain perfectly well. If at the end of fifteen days the hogs receiving both kinds of serum are well, the potent serum is considered to be of standard strength. Should these serum simultaneous hogs (the hogs receiving both the virulent and potent serum) become sick and die of cholera, then the potent serum is considered to be low in potency and is either discarded or larger doses are recommended for field use. It is always advisable to use more than one serum simultaneous hog in testing potent serum as the individual susceptibility of hogs to cholera varies to a great extent.

Where only one animal is used in the testing of potent serum the hog may possess such a high degree of natural immunity that a favorable test could be obtained with a serum below the standard in potency. Because of the impossibility of knowing how much natural immunity a hog has it is advisable to use several pigs in the test. When this is done the chances of making an inaccurate test have been reduced, and consequently less danger of sending out a potent serum below the standard.

In order to lessen the expense of manufacture, efforts have been directed towards other methods of standardizing serum. Giltner of Michigan has shown that potent serum possesses agglutinating properties for *B. cholera suis*. If this could be used as a measure of its efficiency in field work there would be a marked economy in cost of manufacture as well as an important saving of time. With this point in view the testing of potent serum by the agglutination of *B. cholera suis* was undertaken.

DETERMINING POTENCY OF SERUM BY THE AGGLUTINATION OF *B. CHOLERA SUIS*

The strain of *B. cholera suis* used for the agglutination experiment was isolated from the kidney of hog No. 177. This hog was one used in the production of virulent serum and was killed nine days after being injected with hog cholera virus. The lesions were good and typical of hog cholera, as shown by the following autopsy:

Hog No. 177. House M. Killed August 9, 1910:

Autopsy: Well flushed along ventral surface of body. Right inguinal gland normal. Left inguinal gland slightly congested. Petechiae on mucous surface of bladder. Liver pur-

plish but firm. Spleen with blackened hemorrhagic spots. Left kidney badly congested, blackened in appearance and innumerable petechiae on cortex and medulla. Right kidney ditto, but not opened, being used for culture material. Marked congestion and some ulceration in region of ileo-cecal valve. Lungs edematous in interlobular spaces; surface covered with petechiae.

This micro-organism isolated from the kidney of hog No. 177 was labelled 177-k and was undoubtedly *B. cholerae suis* as it corresponded to the description of that organism both culturally and morphologically. One-tenth of a cubic centimeter of a twenty-four culture of 177-k killed a rabbit in eight days. Thus it can be readily seen that this ration of *B. cholerae suis* was highly virulent for rabbits, a characteristic highly desired in the agglutination experiments.

The emulsion of *B. cholerae suis* used in this experiment was made in the following manner. Agar slant cultures, twenty-four hours old, were washed with 4cc of sterile carbol-salt solution, all the growth being removed by the aid of a platinum loop. The carbol-salt solution was made by adding .5 per cent carbolic acid to normal salt solution. The growth was then killed by being exposed to a temperature of 60 degrees Centigrade in a waterbath. This fluid was then filtered through cotton as well as through a filter paper to obtain an even distribution of the organisms. It was then diluted with carbol-salt solution until an emulsion was obtained that was slightly cloudy. Several dilutions were made varying from 1-15 to 1-40. After several tests the dilution of 1-15 was found to give the most satisfactory results and was the dilution used continually throughout the experiment. In order to avoid confusion this emulsion will hereafter be designated as the "test fluid."

The dilutions of potent serum and virulent blood with the test fluid were made as follows: Three c.c. of the test fluid were placed in each of several test tubes. The amount of serum needed to make the corresponding dilutions of 1-100, 1-500, 1-1000, etc., was then added to each of these tubes. In every test a check was carried for the sake of comparison with the various dilutions. In no case was there ever an agglutination in the check tubes. The lower dilutions usually gave a very pronounced agglutination, suspended clumps being easily seen. With higher dilutions it was necessary to shake the tubes in order to ascertain whether or not there was any agglutination. The dilutions were incubated at 37.5 Centigrade and the determinations made at the end of forty-eight hours. Any reaction obtained after that time was not taken into

consideration. When the experiment was first started the dilutions ran no higher than 1-1500 but later they were made as high as 1-1800 and in a few cases 1-10,000 as a routine procedure.

Agglutination Test of Potent Serum, 1910 and 1911

Hog No.	Bleeding	Dilution													
		1-100	1-300	1-500	1-800	1-1000	1-1200	1-1500	1-2000	1-2500	1-3000	1-4000	1-6000	1-8000	1-10,000
1286th		A	A		A		A							
1287th	A	A	A		A									
†128Killing			B		B									
*128Killing			B		B									
1752nd	A	A	B		B									
1753rd	A	A	B		B		B							
2163rd	A	A	A		A									
2351st	A	A	A		A									
2352nd	A	A	A		A									
2353rd	A	A	A		A									
†2354th			B		B									
†2355th			B		B									
*2355th			B		B									
*235Killing		B	B		B									
2541st	A	A	A		A		A		A					
2542nd	A	A	A		A		A		A					
2543rd			A		A									
*254Killing		B	B		B									
†3432nd	B		B		B									
3552nd	A		A		B									
3553rd			B		B									
3751st			B		B									
3761st	A		A		A									
3762nd	A		A		A		A							
3763rd			A		A									
3771st	A		A		A		A							
3772nd			B		B									
3782nd	A		A		A									
3783rd			A		A									
4141st			A		B									

An A indicates a positive reaction; B a negative reaction; a blank space indicates that no dilution was made.

† Test fluid from *B. cholerae suis* No. 177-L.

* Test fluid from *B. cholerae suis*. No. 17.

The first serum tested by the agglutination method was on October 24, 1910, the test fluid being made from the first

or second generation of *B. cholerae suis*. No. 177-k. This test fluid on the whole gave satisfactory results. By December 14th the original test fluid being exhausted, a new test fluid was made up from an organism isolated from the liver of hog No. 177, labelled 177-L, and which corresponded in every way with 177-k. For some unknown reason none of the serum tested with the test fluid No. 177-L gave any reaction, that is, did not produce agglutination. Test fluid 177-L was used on the 4th bleeding No. 235, the killing of No. 128, the 5th bleeding of No. 235, and the 2nd bleeding of No. 343. Since no reaction was obtained with the 177-L test fluid, it was thought that perhaps this culture had passed through too many generations and had lost its virulence. Consequently organism No. 177-k was injected into a rabbit and recovered after the usual methods and was labelled No. 17. A test fluid made from organism No. 17 was used on the killing of No. 128, the 5th bleeding of No. 235, the killing of No. 235 and the killing of No. 254 but no reaction whatever was obtained.

In the fall of 1911 the agglutination experiments were continued. A test fluid was made up from a bacillus isolated from the kidney of hog No. 346, whose cultural and morphological characteristics corresponded with those of *B. cholerae suis*. Its virulence was tested on a rabbit and found to be satisfactory. The experiment this year was not satisfactory, for in most cases no reaction was obtained. Even in those serums giving a positive reaction, the result was not satisfactory in that the period for obtaining the reaction in almost every case extended beyond forty-eight hours.

The following table shows the results of these tests forty-eight hours after being made.

Choosing the dilution of 1-1500 as an arbitrary standard for demonstrating potency, if this method is satisfactory, we should be able to make the following deductions from the above tabulations:

Potent		Non-Potent		Doubtful	
Hog	Bleeding	Hog	Bleeding	Hog	Bleeding
128	6th and 7th	128	Killing	376	3rd
216	3rd	175	2nd and 3rd		
235	1st, 2nd, 3rd	235	4th and 5th		
254	1st, 2nd, 3rd		Killing		
214	1st, 2nd, 3rd	254	Killing		
376	1st and 2nd	355	2nd and 3rd		
377	1st	375	1st		
378	2nd and 3rd	377	2nd		
		414	1st		

As a result of laboratory and field tests of the above serums the following results were obtained:

All of the bleedings which might be judged potent from the agglutinating reactions proved to be satisfactory in the work in the field with the exception of the second and third bleedings of No. 254, from which no report was received. Of the serums which from the above table the agglutinating reactions would show to be non-potent the second and third bleedings from hog No. 175, the second bleeding from hog No. 343, and the first bleeding from hog No. 375, were shown by laboratory tests to be non-potent and were discarded. From the serums from hogs No. 235 and 254 no report was received from the field trials. The remaining serums, that is the different bleedings enumerated from hogs Nos. 128, 355, 377 and 414, all proved satisfactory on field trials.

The serum from hog No. 276 which by the agglutination reaction is placed in the doubtful column proved satisfactory in the field.

While admitting the force of Giltner's argument that confusion must surely result from the comparison of the results of an easily controlled test as the agglutination reaction, with a very elastic standard of biological testing such as is used in the laboratory or field, the object of this experiment was to test the agglutination reaction as a measure of potency in the field.

The results obtained would therefore indicate that there is no constant relationship between the agglutinating reaction of immunizing serum for *B. cholera suis* and its potency as shown by actual field trials. This cannot then be used as a method of standardizing serums for commercial purposes.

THE VALUE OF SALT SOLUTION IN HYPERIMMUNIZATION

The use of a normal salt solution recovered from the intraperitoneal cavity of hogs killed with acute cholera has been used for hyperimmunization purposes in several states. Several hours before these sick hogs are killed they are injected intra-peritoneally with a suitable quantity of a normal salt solution which has been heated to body temperature. After the hogs have been killed, as much of the salt solution as possible is recovered, care being taken to avoid contaminations of any sort. It is the supposition that this injected normal salt solution will absorb a sufficient amount of the cholera virus to make it fit for hyperimmunization. A salt solution recovered from a cholera hog in this manner should, theoretically, be as virulent as the blood of the animal. If this is true it is then possible to have a larger number of hyperimmunes for the

manufacture of potent serum. This work was taken up at this station in the fall of 1911 and satisfactory results were obtained in most cases.

The use of the salt solution was conducted as follows: At least two hours before killing, hogs sick with acute cholera were injected with about 1200 cc. of a normal salt solution. The salt solution was heated to body temperature, the point of injection shaved, cleansed and disinfected and the salt solution injected into the abdominal cavity, the whole operation being conducted with as much care as possible. After the animal was killed, the salt solution was siphoned off into a sterile flask and was later mixed with virulent blood for hyperimmunization. This mixture of virulent blood and salt solution was always tested for virulence upon three pigs, two of which received 2cc of the mixture while the third animal served as a control. The salt solution recovered from hogs Nos. 352 and 353 was tested without being mixed with virulent blood. The salt solution recovered from No. 353 had been in the abdominal cavity for two hours. It proved to be virulent, for the hogs receiving it, Nos. 363 and 364 were killed within fifteen days, which is the required length of time for a virus of standard strength, the post mortem appearances being as follows:

No. 363, slightly flushed on belly and buttocks. Inguinal glands slightly congested. A few petechiae on the cortex of left kidney. Right kidney the same. A few petechiae on the outer surface of colon. Ileo-cecal valve slightly ulcerated and congested. Several collapsed areas on the lungs.

No. 364, flushed on throat, starting to flush on the belly. Inguinal glands congested. Hemorrhagic spots on the liver. Spleen somewhat friable. A few petechiae on the cortex of both kidneys. Several collapsed areas on the lungs. An ulcer on the ileo-cecal valve 3 cm. in diameter.

The control used in this lot proved to be immune for some unknown reason. With the salt solution recovered from hog No. 352, hogs Nos. 357 and 358 received 2cc. of the solution and died in eleven days. They had excellent lesions of cholera and there was no doubt of the virulence of the salt solution as shown by the following autopsies:

Hog No. 357, slightly flushed along throat, sheath of penis and buttocks. Inguinal glands congested. Many petechiae on the cortex of both kidneys. A few petechiae on the mucous coat of bladder. Many hemorrhagic spots on the lungs. Hepatic, mesenteric and lumbar glands congested. Ileo-cecal valve ulcerated.

Hog No. 358, badly flushed along throat, belly and but-

tocks. Inguinal glands congested. Mucous coat of bladder congested. Ileo-cecal valve ulcerated. Areas of collapse and hemorrhagic spots on the lungs.

With one exception the virulent hogs, those receiving virulent blood plus salt solution or just salt solution alone, either died or were killed for acute cholera within 15 days. Hog No. 403 lived 21 days, six days longer than he should, but this can be accounted for in the natural variation in the susceptibility of hogs to attacks of cholera. The controls for the most part were killed in the right length of time for a standard virus which is in the neighborhood of twenty-one days. However, with our work there was always a tendency of control hogs taking on the chronic type of cholera, both when injected with virulent blood alone and when it was mixed with salt solution recovered from cholera pigs. With hogs Nos. 389 and 419, the length of time elapsing from the injection until the animals were killed was so long that the salt solution had been entirely absorbed and none could be recovered.

On the whole the work here with salt solution was considered very satisfactory. The virulence of the recovered salt solution meet the standard requirements and we see no reason why it cannot be used successfully in hyperimmunization. Further investigation as to the maximum amount of salt solution to use and the length of time such solution should remain in the abdominal cavity of sick hogs will throw more light upon the subject and determine its exact value for hyperimmunization.

POTENT SERUM FOR FIELD IMMUNIZATION

Potent serum when sent to the farmer can be used in one of two ways. The hogs may be given only a dose of potent serum, the method being called the serum only method; or they may be given in addition to the potent serum a dose of virulent blood, the latter method being known as the serum simultaneous method. The immunity conferred upon an animal by the use of potent serum alone lasts only for a short period, probably not over three or four weeks. With the serum simultaneous method the immunity lasts from six to nine months, possibly a year. The injection of the virulent blood along with the potent serum creates in the hog an active immunity which permits the animal to withstand an attack of cholera for several months or until it is marketed. Potent serum alone creates no response of the body cells and thus building up no active immunity simply affords the hog protection until the serum disappears.

	Amount of Salt Solution Injected	Amount Re-covered	Length of time from Injection until death of Hog.	Salt Solution How tested Mixed with	Virulence Tested on Hogs Number	Result of Test Hogs, killed or died in Number of Days	Character of Cholera lesions	
363	1700	630	7½	Mixed with Vir. Ser. No. 363	372 373 374	2 days 11 days 17 days	Good Poor Fair	
366	1500	330	7½	Vir. Ser. No. 366	394 395 396	5 days 10 days 15 days	Good Poor Fair	
382	1500	180	22	Vir. Ser. 371, 379, 382	406 407 408	10 days 10 days 43 days	Good Excellent Poor	
383	1500	410	4	Vir. Ser. 374, 383, 390, 393	409 410 ...	12 days 12 days 13 days	Good Excellent Poor	
389	1200	...	7½	
394	1500	460	6	Vir. Ser. 394, 368	403 404 405	21 days 14 days 17 days	Poor Good Fair	Vir. Serum discarded
398	600	100	6	Vir. Ser. 397, 398	424 425 426	10 days 9 days 26 days	Good Good Poor	
404	1500	700	2	Vir. Ser. 402, 404	433 434 435	13 days 12 days 20 days	Fair Good Poor	
406	1200	440	6	Vir. Ser. & Salt Sol. 406, 407	436 437 438	7 days 10 days 20 days	Good Good Poor	
407	1200	200	6	Vir. Ser. & Salt Sol. 406, 407	436 437 438	7 days 10 days 20 days	Good Good Poor	
410	1000	200	24	Vir. Ser. 399, 410	439 440 441	10 days 8 days 22 days	Good Good Poor	
419	1200	...	22	
437	1200	790	5½	Vir. Ser. 434, 437	475 476 477	Immune Immune 14 days	Poor	
352	2000	300	20	Unmixed	357 358 359	11 days 11 days 19 days	Excellent Good Good	
353	2000	850	2	Unmixed	363 365 364	13 days 15 days	Fair Good Good	Proved to be immune

One can readily see the advantage of the serum simultaneous method of vaccinating hogs, nevertheless it is not always employed. For instance, if cholera has already broken out in a herd the remaining hogs are treated with potent serum only, but at the same time they are exposed to the disease by being kept in the same pens or yards with the sick. This exposure acts in the same manner as an injection of virulent blood. Consequently, when cholera appears the well hogs are treated by the serum only method. However, should one desire to protect his herd by vaccination before they had been exposed to cholera, the proper thing to do is to employ the serum simultaneous method because of the much longer period of immunity obtained thereby. For instance, should cholera appear in the neighborhood and a man wants to protect his hogs with potent serum, the serum simultaneous method of injection is used, for the addition of virulent blood would tend to increase the degree of the immunity.

TABLE OF DOSES

Sucking pigs	from 10 to 15 cc
30 to 50 pounds	from 15 to 20 cc
50 to 75 pounds	from 20 to 25 cc
75 to 125 pounds	from 25 to 30 cc
125 to 175 pounds	from 30 to 35 cc
175 to 200 pounds	from 35 to 40 cc
200 to 250 pounds	from 40 to 45 cc
250 to 300 pounds	from 45 to 50 cc
300 to 350 pounds	from 50 to 55 cc
350 to 400 pounds	from 55 to 60 cc

METHOD OF VACCINATION

In vaccinating a drove of hogs with potent serum the first thing to do is to separate them into lots according to their weights. All the hogs of fifty pounds should be in one pen, the hundred pound pigs in another lot, the two hundred pound animals in another pen and so on. This arrangement saves considerable time and trouble, especially in determining the size of the dose. It is sufficient that these weights be only approximate, as there is no danger in giving an excess of the potent serum. In no case should the weights be underestimated in the attempt to economize in the use of serum, for there is danger of cutting the dose down too low. An animal receiving too small a dose is in as much danger as one that has received no serum at all. The dose must in all cases be sufficient to protect the hog from exposure to the most viru-

lent type of the disease. Moreover, giving too small doses is a waste of money and it is much cheaper in the end to use the proper amount of potent serum. The extra cost of serum in giving the correct dosage amounts to a small item compared to the loss which might be incurred through the death of several hogs which had not received enough serum to carry them through a severe exposure.

When cholera has already broken out in a drove of swine before any vaccination has been done the question comes up, which hogs are to be treated and which are not? As already noted potent serum merely protects the hogs from cholera and is in no way a cure for that disease. Hence, animals which show unmistakable signs of being affected with cholera are not to be treated because such treatment proves unsatisfactory. When once the animals are sick with the disease the use of serum has no effect in arresting its progress. If the

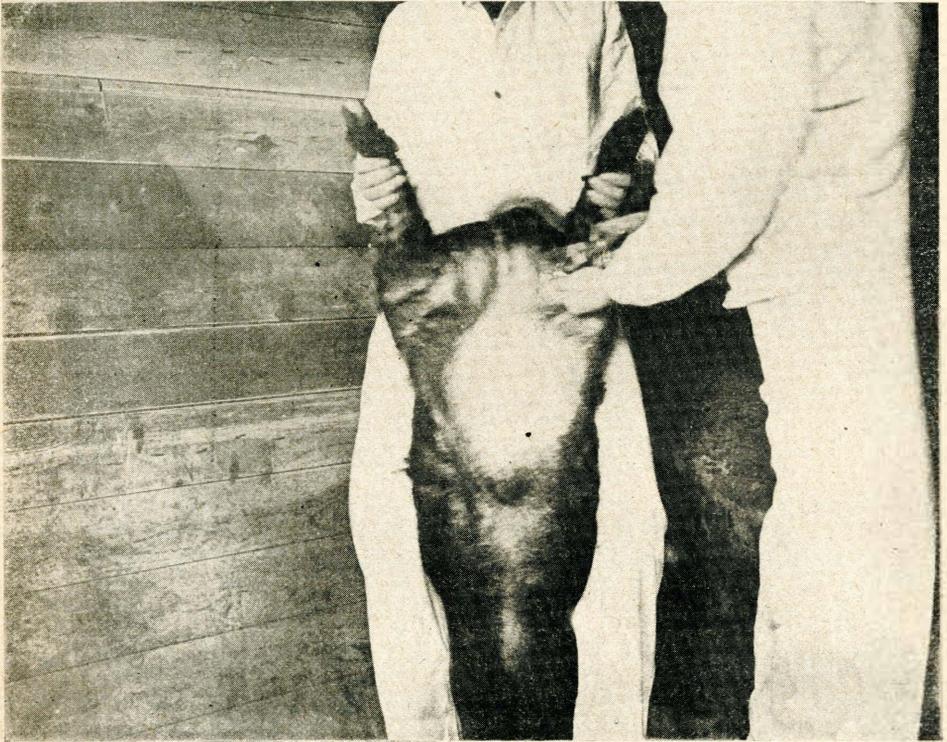


Fig. 5. Showing Method of Holding a Pig While Vaccinating

disease is present in a man's herd the animals which show no symptoms should be separated from the sick. In order to be certain that these apparently well animals are not sick and that the disease is only in its period of incubation, it is advisable to take their temperatures, which should in no case be over 104 degrees Fahrenheit. It must be remembered that usually several days elapse from the time of exposure until cholera can be noticed in the animal, consequently, a hog having a temperature higher than 104 degrees is very suspicious and will very likely be noticeably sick within a short time.

A convenient method of holding small pigs weighing up to a hundred pounds while being vaccinated, is to hold the pig's hind legs in the air with the belly towards the operator, and its head between the legs of the assistant. (See Figure 5.) Larger hogs can be thrown and held or put in a crate during the operation. A large hog can best be vaccinated by tying a rope around its snout and then snubbing it up to a post or tree.

The potent serum should be injected deeply into the flesh on the inside of the left hind leg and the virulent blood on the inside of the right hind leg. This is not strictly essential but it is much better to follow a routine method in order to avoid mistakes. The site of injection should be disinfected with a 4 per cent compound solution of cresol or 5 per cent carbolic acid solution. This is best done by wiping off the skin with a small piece of cotton which has been soaked in one of the above named antiseptic solutions. Where animals become lame for two or three days after injection it is no indication that the serum is "working" or has "taken," but rather that accidental infection has occurred by the introduction of other germs during the operation of vaccination, and that these adventitious micro-organisms have caused the lameness, not the serum.

SANITARY MEASURES

The carcasses of dead hogs should be either buried or burned. Burning is more preferable, as there is no danger of dogs or pigs rooting out the buried animals and thus spreading contagious material about. After an outbreak of cholera all pens and yards should be thoroughly cleaned and disinfected. All manure, litter, and straw should be burned; then a good application of coal tar dip applied. The virus of cholera is extremely contagious and may be carried about by birds, dogs, on the shoes of a man and in other ways which cannot be accounted for. Cholera may also be spread by running water. Frequently where pens are near the bank of

a stream, cholera is spread along the creek or river. When such is the case sanitary measures should be strictly enforced. Infected pens should then be disinfected and all unaffected herds should be vaccinated as a precautionary measure. The pens should be well constructed and made so that they can be cleaned. Proper feeding, plenty of exercise, clean pens and an abundance of sunshine will do a great deal towards protecting hogs from cholera. Disease of any kind flourishes in filth and dirty pens and yards. Therefore, it behooves every breeder of hogs to exercise the best of care and keep his animals healthy and clean. This does not mean that hogs kept under such conditions will not get the cholera, but that they have more chances of avoiding this dreadful disease.

DOES VACCINATION PREVENT HOG CHOLERA

The average hog raiser still regards this as an open question, and this method of prevention in its experimental stages. An extremely conservative estimate of the results obtained by this laboratory shows that ninety per cent of all animals treated have safely withstood the disease. It is frankly admitted that in a few isolated cases the number of animals successfully coming through the outbreak in a herd has been less than this, but in such cases, with the single exception of one herd, treated by the first serum manufactured, the loss is directly attributable to using too small a dosage of the potent serum, or the vaccination of hogs too far advanced in the period of incubation. In this connection attention is also directed to the fact that almost without exception this method of treatment has only been adopted after the disease has already appeared in a herd. All other laboratories and stations concerned in the manufacture and distribution of this serum have reported much the same results.

So far as the experimental aspects of this question are concerned, they are therefore confined to inquiries into new methods of preparing, testing, or using the serum in an effort to lessen the cost to the hog raiser of immunizing his hogs against cholera.

COST OF VACCINATION

Potent and virulent hog cholera serum is distributed approximately at cost, which under the present methods of manufacture is estimated at two cents per cubic centimeter. The cost of vaccination is therefore directly dependent upon the weight of the hogs to be treated and by consulting the table of dosage will be found to vary from twenty to thirty cents for

a small pig to one dollar and a half for a five hundred pound pig. So far as vaccination against hog cholera is concerned therefore it can readily be regarded as a cheap form of insurance policy against this disease, allowing the owner to retain the full number of animals in the herd and to keep them over until fully matured for market.

To intelligently fill requests for serum it becomes necessary to know:

1. The number of hogs in the herd.
2. The approximate weight of the individuals in the herd.
3. Whether or not cholera is present in the herd or simply in the immediate locality.
4. If the disease is present, the number that show any indications of being sick.

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