LEPTOSPIRA POMONA AS A CAUSE OF ABORTION AND NEONATAL MORTALITY IN SWINE.

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SUMMARY.

The effect of Leptospira pomona **infection of sows during pregnancy was studied.**

Four sows infected intramuscularly with a culture of L. pomona **in the second half of pregnancy produced 37 piglets, of which 34 were born dead or died within a few minutes of birth. Four uninfected sows kept under similar conditions produced 35 piglets, of which six were born dead or died within a few minutes of birth.**

The average gestation period in the four uninfected sows was 115 days, whereas in the four infected sows the average period was 95 days. The duration of abortion in the infected sows appeared to be longer than farrowing in the uninfected sows.

There was no clinical evidence of ill health in three of the four sows, but the other showed a temperature reaction and anorexia for a few days.

The infected sows commenced excreting Leptospira in the urine 17, 19, 9 and 10 days after infection and excreted for a period of 5, 7, 83 and 24 days respectively.

Approximately 40% of the dead piglets of the infected sows showed focal necrosis of the liver and excess serous fluid in the body cavities. The remainder were either macroscopically normal or were partly decomposed.

Leptospira were isolated from 12 of 35 piglets from the infected sows.

The diagnosis of porcine leptospiral abortion is discussed. It is recommended that dead piglets be examined by microscopic examination of peritoneal fluid and guinea pig inoculation of tissues.

INTRODUCTION.

Observations on two naturally infected herds of swine and on one experimentally infected sow indicated that *Leptospira pomona* infection may result in the birth of weak or dead piglets (Ryley and Simmons 1954). To our knowledge no further experimental work on swine leptospirosis has been published subsequent to the submission of that paper, although Reinhard (1953) in a review of the subject quoted a personal communication stating that Leptospira had been recovered from aborted porcine foetuses.

The present paper records observations on a further four sows experimentally infected with *L. pomona,* and on four other comparable uninfected sows kept under the same conditions.

METHODS.

Experimental Sows.

Eight maiden sows and a boar of the Large White breed were selected from a brucellosis-tested piggery, after blood samples from these animals were found negative to the agglutination tests for brucellosis and *L. pomona.*

Housing.

The sows were run together until a few days before infection, then separated, each being placed in a concrete pen, 8·5 ft. x 18·5 ft. Nine pens in a row were used, the group selected for infection with Leptospira being separated from the uninfected sows by an empty pen. The uninfected sows were always attended first.

Feeding.

The sows and the boar were fed from about two weeks before mating commenced on a mixture containing:—

	Per cent. (by weight).
Crushed sorghum	18
Crushed wheat	30
Crushed maize	25
Meatmeal (55% crude protein)	12
Dried buttermilk powder (35% crude protein)	3
Livermeal (65% crude protein)	3
Lucerne chaff	5
Bran	2
Ground limestone	1
Salt	1

A fish-liver oil emulsion was added to the mixture to provide approximately 5,000 I.U. of vitamin A per lb.

The ration, which provided approximately 19% crude protein, was mixed weekly. Approximately 5 lb. was fed dry per day to each sow at the beginning of the experiment, but this was gradually increased to 6 lb. at farrowing. Freshly cut lucerne was fed at the rate of 2–3 lb. daily.

Clinical Observations.

The sows were examined daily and rectal temperatures recorded twice daily during the course of the experiment.

Urine Specimens.

Daily urine samples were collected from the infected animals and examined as described previously (Ryley and Simmons 1954). Samples were collected at least once and usually twice weekly from the uninfected sows.

Strain of Leptospira.

The *L. pomona* strain described in our other paper (Ryley and Simmons 1954) was used. Four sows (Nos. 3, 4, 5 and 6) were inoculated intramuscularly with 1·0 ml. of culture from the twenty-fifth guinea pig passage, while the remaining four (Nos. 7, 11, 12 and 40) served as uninfected controls. Both groups were comparable in body weights and dates of service.

The weights of the control sows three weeks before infection date varied between 257 lb. and 297 lb. (average 282 lb.) and of the infected group between 263 lb. and 306 lb. (average 285 lb.).

Agglutination Tests.

Blood samples were collected at intervals varying from 10 days to one month. The sera were kept at –25°C. until the end of the experiment and then examined by the agglutination test (Ryley and Simmons 1954).

Examination of the Piglets.

Farrowing was observed whenever possible and any dead piglets autopsied within 12 hours. A loopful of pleural fluid, peritoneal fluid, stomach content and urine was examined by dark ground illumination. Portion of the lung, liver, stomach content and kidney was ground with sterile sand and saline and 1 ml. of the supernatant inoculated intraperitoneally into guinea pigs of approximately 250 g. weight. Liver and kidney was sown on sheep's blood agar plates to check for *Brucella* spp. Rectal temperatures of the guinea pigs were recorded twice daily for one week; peritoneal fluid of those showing a temperature over 104°F. was obtained by the method of Van Thiel (1948) and examined for Leptospira by dark ground illumination. All guinea pigs were slaughtered after three weeks and their sera submitted to agglutination tests for *L. pomona* and brucellosis.

RESULTS.

Clinical Observations.

The uninfected sows remained healthy throughout the experiment, except sow 11, which had Group C streptococcal mastitis in the posterior unsuckled teats nine days after farrowing. This sow was treated by intramuscular inoculation of 500,000 units of procaine penicillin daily for three days and recovered.

Temperature charts of the four infected sows (Nos. 3, 4, 5 and 6) are shown in Figs. 1-4. Sow 3 showed no abnormality except a slight rise (103·8°F.) on the thirteenth day after inoculation (Fig. 1). Sow 4 also remained normal and showed a rise (104·2°F.) on the day after farrowing (Fig. 2).

Sow 5 had inappetance on the fifth and sixth days after infection and temperatures of 103·7°F. four days after inoculation and 104·0°F. on the afternoon of the day of farrowing.

Sow 6 had two periods of ill health. She showed dullness and anorexia between the fifth and seventh days after infection with an increased temperature between the third and seventh days, reaching a peak of 105·5°F. on the afternoon of the fourth day. Anorexia and a slight ocular discharge occurred on the sixth to seventeenth day after farrowing, with a temperature rise between the sixth and twelfth days. The highest temperature of 105·6°F. was recorded on the afternoon of the seventh day. Appetite gradually returned and was normal on the twentieth day after farrowing.

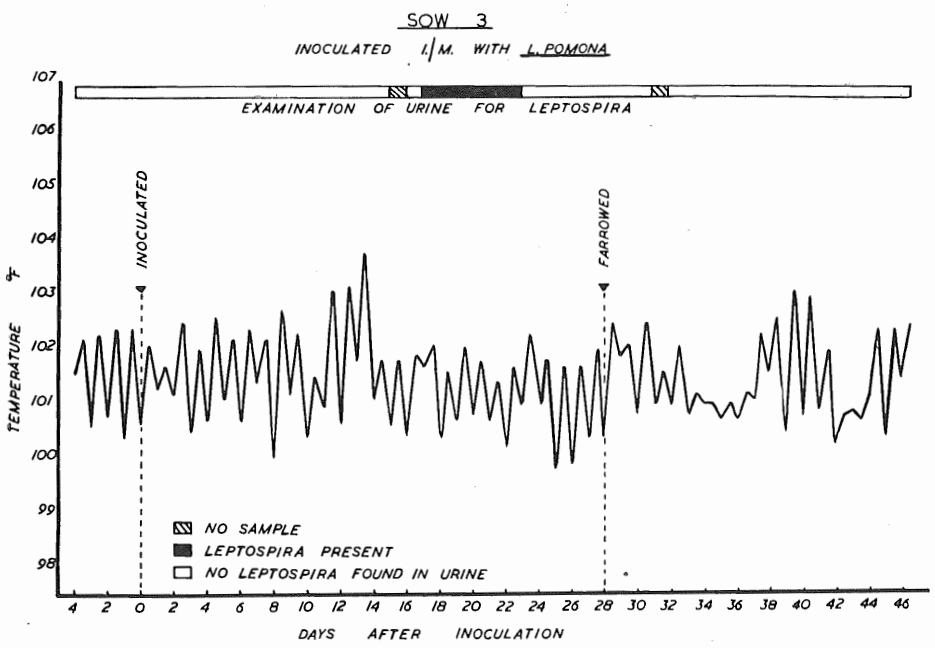

Fig. 1.

Sow 3 Inoculated Intramuscularly with a Culture of *L. pomona*. Rectal temperatures twice daily and results of dark ground examinations of urine for Leptospira.

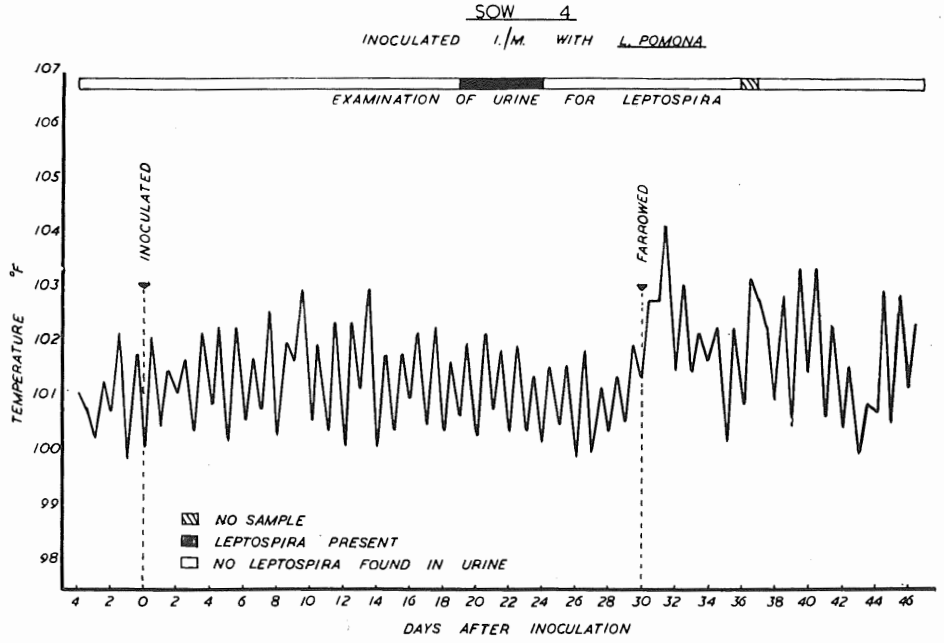

Fig. 2.

Sow 4 Inoculated Intramuscularly with a Culture of *L. pomona*. Rectal temperatures twice daily and results of dark ground examinations of urine for Leptospira.

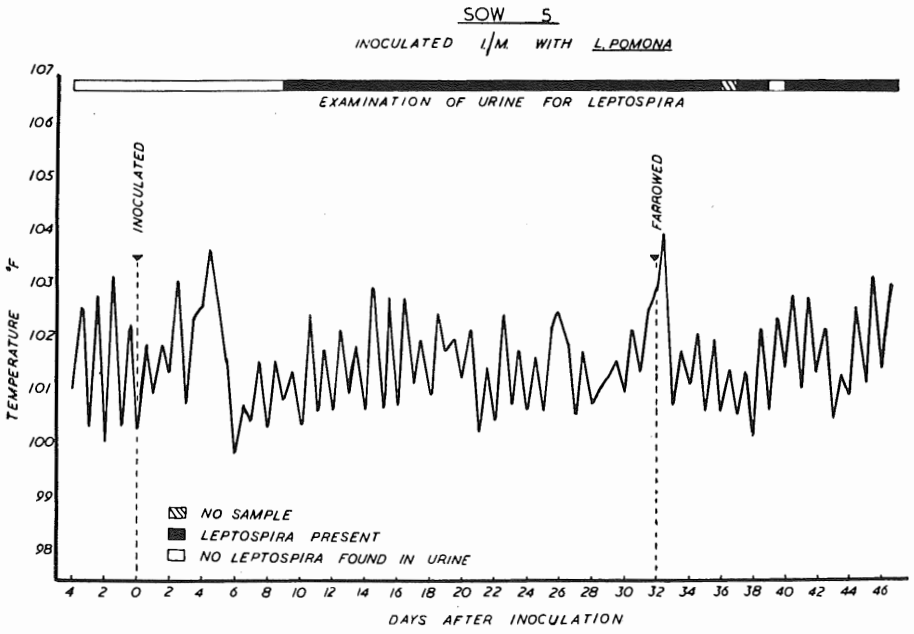

Fig. 3.

Sow 5 Inoculated Intramuscularly with a Culture of *L. pomona*. Rectal temperatures twice daily and results of dark ground examinations of urine for Leptospira.

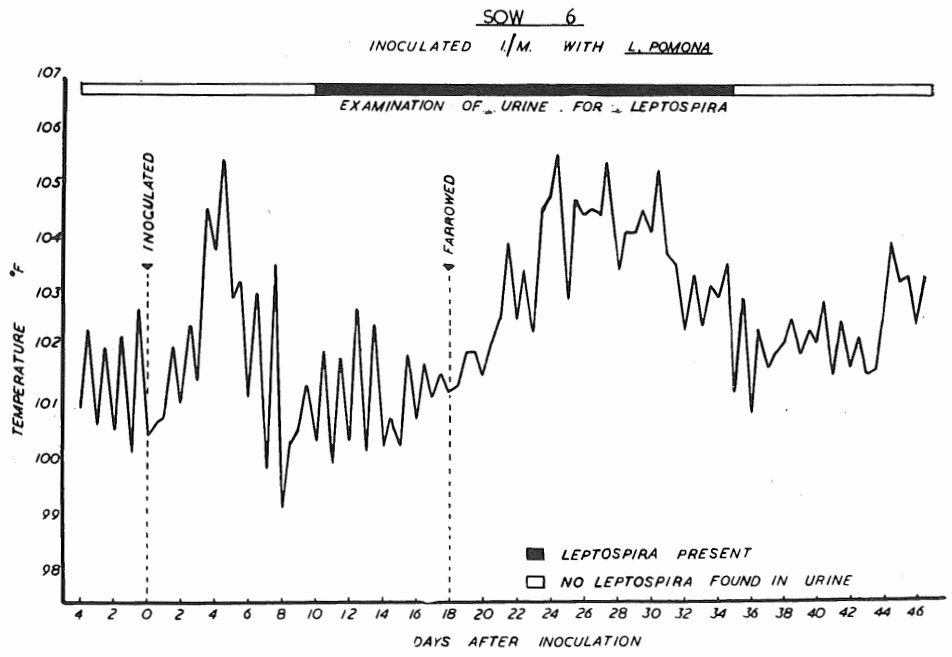

Fig. 4.

Sow 6 Inoculated Intramuscularly with a Culture of *L. pomona*. Rectal temperatures twice daily and results of dark ground examinations of urine for Leptospira.

Urine Examination.

Between 14 and 20 urine samples were collected from each uninfected sow during the experimental period, and all were negative for Leptospira.

Results of urine examinations of the infected sows are given in Figs. 1-4. The commencement, cessation and duration of leptospiruria were as follows:—

Sow No.	Leptospiruria. (days after inoculation).		Duration. (days.)
	Commenced.	Ceased.	
3	17	24	7
4	19	24	5
5	9	92	83
6	10	34	24

Sows 3 and 4 ceased excreting Leptospira five and seven days respectively before aborting. Sows 5 and 6 were excreting them at the time of parturition.

Serology.

The results of the agglutination tests are shown in Table 1.

Table 1.

RESULTS OF AGGLUTINATION TESTS ON SERA FROM UNINFECTED AND INFECTED SOWS.

Sow No.	Dec. 4.	Dec. 10.	Dec. 22.	Jan. 15.	Feb. 15.	March 8.
7	0	0	0	0	0	0
11	0	0	0	0	0	0
12	0	0	0	0	0	0
40	...	...	...	0	0	0
3	0	0	300*	3,000	1,000	300
4	0	0	1,000	300	300	100
5	0	0	1,000	3,000	3,000	3,000
6	0	0	1,000	30,000	10,000	10,000

Key :— * Reciprocals of titres. Read at approximately 50% agglutination.
0 = No agglutination.
... = No sample.

Note :—The infected sows were inoculated on December 11.

Sera from the four uninfected sows showed no agglutination whereas those from the four infected sows had a maximum titre of between 1:300 and 1:30,000. Eleven days after inoculation all infected sows showed a titre of 1:300 or greater. The highest titre (1:30,000) occurred in the sera from sow 6, which showed symptoms of anorexia and hyperpyrexia. It will be noted that two of the four infected sows, Nos. 3 and 4, showed titres of 1:300 and 1:1,000 respectively before Leptospira were found in the urine. The other two sows were excreting Leptospira before the first post-infection bleeding.

Details of Farrowing.

The gestation periods and number of piglets born are shown in Table 2.

Table 2.

PARTICULARS OF GESTATION PERIODS AND LITTERS OF UNINFECTED AND INFECTED SOWS.

Sow No.	Date of Service.	Date of Infection.	Date of Parturition.	Gestation Period. (days)	Number of Piglets.		
					Born.	Born dead.	Survived 2 days.
7	Oct. 9	Nil	Feb. 1	115	8	0	8
11*	Oct. 10	Nil	Feb. 3	116	11	3	4
12	Sept. 15	Nil	Jan. 8	115	2	0	2
40	Oct. 25	Nil	Feb. 16	114	14	3	11
3	Oct. 12	Dec. 11	Jan. 10	90	13	13	0
4	Oct. 11	Dec. 11	Jan. 11	92	6	6	0
5	Oct. 14	Dec. 11	Jan. 13	91	10	10	0
6	Sept. 14	Dec. 11	Dec. 30	107	8	5	1

* Litter with congenital defects.

Three of the uninfected sows farrowed within three hours and the fourth (sow 40) farrowed 13 of the 14 piglets during 90 minutes and the last piglet within 10 hours.

In the infected group farrowing occupied 22 hours in sow 4, was less than 16 hours in sow 3 (overnight), between 4 hours and 8 hours in sow 5 and 6-12 hours in sow 6. Sows 3, 4 and 5 aborted, while in sow 6 pregnancy terminated a week before the due date.

The four uninfected sows produced 35 piglets, of which six were dead or died within a few minutes of birth. One of these six was a partially resorbed mummified foetus (Table 3). All the piglets produced by sow 11 showed the congenital defect "kinky tail" (Donald 1949), which will be described in a later paper by Ryley and Melville. Of the eight live piglets in this litter, five had cleft palates and were either destroyed or died within the first three days because of their inability to suckle. One piglet from sow 7 died from overlying on the fourth day.

The four infected sows produced 37 piglets, of which 34 were dead or died within a few minutes of birth (Table 4). Of the three live piglets, one died after five hours, another within 48 hours and the third was weaned at eight weeks. Serological evidence and urine examinations indicated that this piglet was not infected.

Table 3.

BIRTH WEIGHTS, SURVIVAL, PATHOLOGY AND BACTERIOLOGY OF PIGLETS FROM UNINFECTED SOWS.

Sow No.	Piglet No.	Birth Weight. (lb.)	Survival.	Pathology.	Bacteriology.
7	1	2·3	Weaned	...	...
	2	2·4	Weaned	...	...
	3	2·6	Weaned	...	...
	4	2·3	Weaned	...	...
	5	1·9	3 days	Trauma and ruptured liver	—
	6	2·6	Weaned	...	...
	7	2·5	Weaned	...	...
	8	2·8	Weaned	...	...
11	9	1·5	2 days	Stomach empty. Hare lip and cleft palate	—
	10	2·1	Weaned	...	...
	11	2·3	Weaned	...	...
	12	1·9	Weaned	...	...
	13	2·1	3 days	Stomach empty. Hare lip and cleft palate	—
	14	2·3	5 hours	Destroyed. Hare lip and cleft palate	—
	15	1·9	5 hours	Destroyed. Hare lip and cleft palate	—
	16	1·5	5 hours	Destroyed. Hare lip and cleft palate	—
	17	1·8	0	Hare lip and cleft palate	—
	18	0·9	0	Decomposed. Hare lip and cleft palate	—
	19	0·3	0	Mummified. Hare lip and cleft palate	...
12	20	3·2	Weaned	...	...
	21	2.5	Weaned	...	...
40	22	1·8	Weaned	...	...
	23	2·7	Weaned	...	...
	24	1·9	Weaned	...	...
	25	1·9	Weaned	...	...
	26	2·4	Weaned	...	...
	27	1·2	0	Lungs atelectic. N.A.D.	—
	28	1·8	Weaned	...	...
	29	1·5	Weaned	...	...
	30	1·9	0	Lungs inflated. N.A.D.	—
	31	1·7	Weaned	...	...
	32	1·5	Weaned	...	...
	33	2·3	Weaned	...	...
	34	1·9	0	Lungs atelectic. N.A.D.	—
	35	2·3	Weaned	...	...

Key :—N.A.D. = No abnormality detected.
... = No examination.
— = Negative.

Pathology of Piglets.

No significant abnormalities were seen in any piglets from the uninfected sows, except for the congenital defects in the litter of sow 11. Only two of the dead piglets (Nos. 18 and 19 from sow 11) showed evidence of partial decomposition at birth.

Twenty-four of the 36 piglets from the infected sows showed either focal necrosis of the liver with excess clear yellow pleural and peritoneal fluid or blood-stained fluid throughout the subcutis and in the pleural and peritoneal cavities. The necrotic foci varied in number, were irregular in shape and 1-4 mm. in diameter (Fig. 5). Histologically the livers with macroscopic lesions showed foci of acute necrosis, with distinct margins and almost no cellular reaction. There was no significant abnormality in 10 of the piglets and two were mummified. Thirteen piglets showed evidence of death having occurred some time prior to birth (Table 4).

Some of the foetal membranes from the infected sows showed thickening and oedema of the chorio-allantois and an irregular whitish deposit on the uterine surface of the chorion. Membranes of the decomposed piglets were necrotic and greyish-brown in colour. Some decomposed membranes had red or brown raised circular areas 1-4 mm. in diameter.

Table 4.

BIRTH WEIGHTS, SURVIVAL, PATHOLOGY AND BACTERIOLOGY OF PIGLETS FROM INFECTED SOWS.

Sow No.	Piglet No.	Birth Weight. (lb.)	Survival.	Pathology.	Bacteriology.
3	1	1·0	0	N.A.D.	+
	2	0·7	0	N.A.D.	...
	3	1·3	0	Focal necrosis of liver. Excess serous fluid	+
	4	1·3	0	Liver orange and friable	−
	5	1·0	0	Focal necrosis of liver. Excess serous fluid	+
	6	0·5	0	Focal necrosis of liver	+
	7	0·9	0	N.A.D.	−
	8	1·4	0	N.A.D.	−
	9	1·1	0	N.A.D.	−
	10	0·8	0	Decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	−
	11	0·7	0	Decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	−
	12	0·4	0	Decomposed. N.A.D.	−
	13	0·2	0	Partially resorbed mummified foetus	...

Table 4—*continued.*

Sow No.	Piglet No.	Birth Weight. (lb.)	Survival.	Pathology.	Bacteriology.
4	14	1·2	0	Decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	—
	15	1·5	0	Inflated lungs. N.A.D.	—
	16	1·5	0	Injuries by sow after death	—
	17	1·5	0	Inflated lungs N.A.D.	—
	18	1·6	0	Decomposed. Focal necrosis of liver. Blood-stained fluid in peritoneal and pleural cavities and stomach	—
	19	1·3	0	N.A.D.	—
5	20	0·3	0	Mummified	—
	21	0·7	0	Slightly decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	—
	22	0·7	0	Slightly decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	—
	23	0·8	0	Focal necrosis of liver. Excess serous fluid	+
	24	0·7	0	Focal necrosis of liver	+
	25	0·6	0	Focal necrosis of liver. Excess serous fluid	+
	26	0·8	0	Focal necrosis of liver. Excess serous fluid	+
	27	0·6	0	Focal necrosis and reddish depressed areas in liver. Excess serous fluid	+
	28	0·8	0	Excess serous fluid	+
	29	0·8	0	Focal necrosis of liver. Excess serous fluid	+
6	30	1·8	0	Decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	—
	31	1·6	0	Decomposed. Focal necrosis of liver. Blood-stained fluid in pleural and peritoneal cavities	—
	32	1·7	5 hours	Focal necrosis of liver. Excess serous fluid	+
	33	1·5	0	Focal necrosis of liver. Excess serous fluid. Petechiae on epicardium	—
	24	1·4	0	Decomposed. Blood-stained subcutaneous, pleural and peritoneal fluid	—
	35	2·4	0	Decomposed. Focal necrosis of liver. Blood-stained peritoneal and pleural fluid	—
	36	1·8	1 day	Focal necrosis of liver	—
	37	2·1	Weaned	...	...

Key :—N.A.D. = No abnormality detected.
... = No examination.
— = Negative.

Bacteriology of Piglets.

No Brucella were isolated from any of the dead piglets by culture and guinea pig inoculation.

No Leptospira were isolated from the dead piglets of the uninfected sows, but 12 of the 35 piglets examined from the infected sows were found to be infected. All these 12 piglets were shown to be infected by guinea pig inoculation, but only nine of them were positive on direct examination. Of the nine piglets showing Leptospira on direct examination, all showed

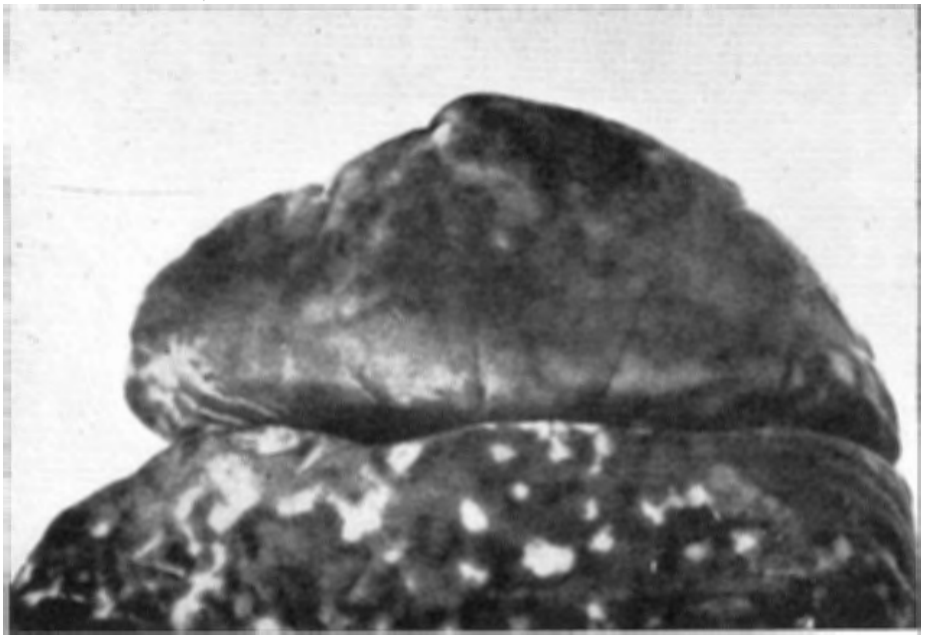

Fig. 5.

Portion of the Liver of Piglet No. 23 (Infected) Showing Focal Necrosis.

Leptospira in the peritoneal fluid, 7 in the pleural fluid, 4 in the urine and one in the stomach contents. Sera from the infected guinea pigs agglutinated a known strain of *L. pomona*. Leptospira were not recovered from any of the piglets of sow 4 (Table 5).

Dark ground examination of the foetal membranes from one sow was positive, from another negative; the membranes from the other two sows were not examined.

Table 5.

RESULTS OF EXAMINATION FOR LEPTOSPIRA OF DEAD PIGLETS FROM UNINFECTED AND INFECTED SOWS.

Sow No.	Number of Piglets Examined.	Number Infected.		
		Microscopic Examination.	Guinea Pig Inoculation.	Total.
7	1	0	0	0
11	0	0	0	0
12	7	0	0	0
40	3	0	0	0
3	12	2	4	4
4	6	0	0	0
5	10	6	7	7
6	7	1	1	1

DISCUSSION.

Four sows infected with *Leptospira pomona* in the second half of pregnancy produced 34 stillborn piglets out of 37 piglets born. Four uninfected sows kept under similar conditions produced six stillborn piglets out of 35 piglets born.

Three of the four infected sows aborted 3-4 weeks before full term, whilst the other farrowed one week before the anticipated date. The four uninfected sows farrowed at term. Protracted parturition in the infected sows is probably not specific for leptospiral abortion, as it also occurs in Brucella infection (Thomsen 1935).

Two infected sows showed no clinical evidence of the disease, one showed a transient inappetance which could have been overlooked in communal feeding, while the fourth was dull, fevered and did not eat for some days. The latter did not show icterus or haemoglobinuria commonly associated with leptospirosis in other species of animals. There was no clinical evidence of meningeal involvement, although this is often present in human cases (Van Thiel 1948).

The piglets from the infected sows were normally developed, but because of the shorter gestation period their weights were lower than those from the uninfected sows. The state of some of the foetuses at birth would indicate that death had occurred *in utero* several days prior to expulsion. As only one piglet from an infected sow survived we have insufficient data on the effect of leptospirosis on the growth of piglets. However, it was observed that this piglet and two others born alive were not active and did not try to suckle immediately after birth.

The presence of focal necrosis of the liver in stillborn piglets shows clearly that they were infected *in utero*. The interval between intra-uterine death and expulsion appeared to determine whether these lesions were present and had some bearing on the chance of isolating the organism. Piglets that died at or just prior to birth often showed this lesion and Leptospira were present, whereas those that had died earlier showed only partial decomposition and we were unable to recover Leptospira from any of them. In contrast with normal foetal membranes, those produced by infected sows were either thickened and oedematous or brown and necrotic. As two of the sows showed no significant temperature rise it is suggested that abortion is not due to pyrexia but to infection of the foetus and its membranes.

The period of leptospiruria varied considerably, ranging from 5 days to 83 days. This is much shorter than that recorded by Schmid and Giovanella (1947) for young pigs. It would be interesting to study the effect of age and pregnancy on the duration of leptospiruria. In two of our infected sows the agglutination titre was positive before the organism appeared in the urine.

Our present knowledge of herd history and clinical symptoms in leptospiral abortion is not sufficient to permit differentiation from other causes of abortion. Presumptive evidence can be obtained by serum agglutination tests and dark ground examination of urine from the breeding sows. The interpretation of serological reactions is difficult, as a positive reaction may occur in recovered animals. We consider that a sow excreting Leptospira in the urine in the pregnancy may produce an unsatisfactory litter, but that absence of Leptospira in a urine sample does not ensure that the litter will be free of the effects of leptospiral infection of the sow.

Examination of the dead piglets or foetuses is the best method of diagnosis. As not all dead piglets in a litter may show the lesions and/or harbour the organism, all those born dead should be examined. However, in some litters Leptospira cannot be detected in any of the piglets. Mummified foetuses, which may also occur in litters from uninfected sows, are unsuitable for examination.

The piglets should be examined for evidence of focal necrosis of the liver, and the peritoneal and pleural fluid should be examined under dark ground illumination. As the epidemiology of *L. pomona* infection may differ from that of other species of Leptospira, it is desirable that the strain be isolated and identified. Guinea pig inoculation is more satisfactory for isolation than culture of material because of the frequent presence of bacterial contaminants.

Our experimental results indicate that economic loss associated with the adult pig is small, but that loss associated with the birth of dead piglets can be considerable.

There is a danger of infection of man, not only from infected urine, but also from the handling of foetuses or membranes.

Although our experiments showed that sows infected in the second half of pregnancy may abort and produce dead piglets, we do not know whether this will also occur if the sows are infected before mating, during early pregnancy or in very late pregnancy.

The four infected sows used in this experiment have now been re-mated to the same boar to study subsequent breeding performance.

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