A Serological Survey of Leptospirosis in Prince Edward Island Swine Herds and its Association with Infertility

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ABSTRACT

A serological survey was undertaken to determine the prevalence of leptospirosis, and to investigate associations between leptospiral antibody titers, and herd measures of reproduction. Production records and leptospirosis serology were analyzed for 25 slaughter hogs from each of eleven randomly sampled farrow-finish operations on Prince Edward Island. The effect of selected leptospiral serovars on nonproductive sow days per parity (NPSD/P) and the proportion of pigs born dead was evaluated. The four most common serovars to which antibodies were detected were *Leptospira icterohaemorrhagiae*, *L. bratislava*, *L. autumnalis* and *L. pomona*, with respective prevalences of 57.1%, 35.1%, 3.4% and 1.5% of PEI slaughter hogs. None of these serovars was associated with increased frequency of stillbirths ($p > 0.05$). However, farms with a higher prevalence of *L. bratislava* antibody titers tended to have more infertility, as measured by NPSD/P ($r = 0.738$, $p = 0.036$ with Bonferroni adjustment). Also, farms with *L. pomona* antibody titers had higher NPSD/P than farms without *L. pomona* antibody titers ($p = 0.0008$ with Bonferroni adjustment). There was no association between NPSD/P and antibodies to either *L. autumnalis* or *L. icterohaemorrhagiae* ($p > 0.05$).

RÉSUMÉ

Une enquête sérologique a été effectuée afin de déterminer la prévalence de la leptospirose et pour rechercher des relations possibles entre les titres d'anticorps et certaines données relatives aux performances de reproduction. La collecte des données et les analyses sérologiques pour la leptospirose ont été effectuées à partir de 25 porcs en âge d'être abattus, provenant de 11 élevages de type naisseur-finisseur et choisis au hasard, sur l'Île-du-Prince-Édouard. La relation entre la présence de certains sérovars de leptospires, le nombre de jours non-productifs par truie par année selon la parité (NPSD/P) et la proportion des porcelets mort-nés, a été évaluée. Les sérovars contre lesquels des anticorps ont été décelés le plus fréquemment ont été *icterohaemorrhagiae*, *bratislava*, *automnalis* et *pomona*, avec des prévalences respectives de 57,1 %, 35,1 %, 3,4 % et 1,5 %. Aucune corrélation n'a pu être établie entre l'un ou l'autre de ces sérovars et la fréquence de mortinatalité ($p > 0.05$). Toutefois, dans les fermes ayant une prévalence élevée de séro-réaction contre le sérovar *bratislava*, on remarquait plus d'infertilité, selon le NPSD/P ($r = 0,738$, $p = 0,036$, ajustement selon Bonferroni). De plus, les fermes possédant des sujets séro-réacteurs au sérovar *pomona* avait un NPSD/P plus élevé que dans les fermes sans séro-réacteurs à ce sérovar ($p = 0,0008$, avec ajustement selon Bonferroni). Aucune corrélation n'a pu être mise en évidence entre le NPSD/P et la présence d'anticorps contre les sérovars *automnalis* ou *icterohaemorrhagiae* ($p > 0,05$). (*Traduit par Dr Robert Higgins*)

INTRODUCTION

Leptospirosis is a zoonotic disease that causes a persistent infection in swine leading to reproductive problems such as abortions, infertility, stillbirths and weak piglets. All pathogenic serovars are classified as *Leptospira interrogans* (1). Many pathogenic serovars have been recovered from swine (2–5), although the clinical significance of these findings can be difficult to interpret.

The objectives of this study were to determine the serological prevalence of leptospirosis in Prince Edward Island (PEI) slaughter hogs, and to investigate associations between leptospiral titers and herd measures of reproduction.

MATERIALS AND METHODS

A random sample of the farrow-finish farms (11 farms) described in a previous paper (6) were asked to participate in a serological survey for leptospirosis, as well as maintain detailed production records for one year.

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During July and August of 1988, blood was collected from 25 slaughter hogs from each farm. This sample size was calculated to allow detection of 5% prevalence (2) within each farm, 95% of the time. The serum samples were tested for *Leptospira interrogans* antibodies to the following serovars (strain in parentheses): *pomona* (pomona), *hardjo* (hardjoprajitno), *canicola* (hond, Utrecht), *copenhageni* (M20), *icterohaemorrhagiae* (RGA), *grippotyphosa* (Andaman), *tarassovi* (perepelicin), *australis* (ballico), *bratislava* (Jez brat), *autumnalis* (Akiyama A), *ballum* (Mus 127), *hebdomadis* (hebdomandis), *javanica* (poi) and *bataviae* (Van Tienen). The test used was the microscopic agglutination test (7). A titer of 1:100 or greater was considered positive. A farm was classified positive if one or more animals tested positive.

For analysis, all data were entered into a microcomputer based data management software package. Descriptive statistics, correlations and *t*-tests were calculated using "Minitab" (8). To determine if the two most common serovars were reacting to the same antigen, Kappa (9), and McNemar's chi-square (10) were calculated.

RESULTS

All producers agreed to participate; there was no loss to follow-up. Prevalence results are found in Table I. There were no reactions to *L. australis*, *L. ballum*, *L. bataviae*, *L. grippotyphosa*, *L. hebdomadis*, *L. tarassovi*. Titers were generally low (1:100 or 1:200). Only 16 hogs had a titer of 1:400 to *L. bratislava* and/or *L. icterohaemorrhagiae*. One hog had a titer of 1:3200 to *L. bratislava*. Antibodies to a total of eight serovars were detected with all herds having *L. bratislava* and *L. icterohaemorrhagiae* titers. With the exception of those two serovars, all other antibodies were of low prevalence (< 4%). Since 75% of the serum samples with a titer to *L. bratislava* also had a titer to *L. icterohaemorrhagiae*, the possibility of a cross-reaction (5,11) was investigated. Kappa was 0.23 (p = 0.000) (12); although there was 60% agreement betweeen the two serovars observed, 48% would have been expected due to chance. As well, results were put in a two by two contingency table which had a McNemar's chi-square of 32.53 (p = 0.000).

TABLE I. Prevalence of leptospiral titers in porcine sera on Prince Edward Island

Serovar	Sera (N = 268) # Seropositive	Sera (N = 268) % Seropositive	Herds (N = 11) # Seropositive	Herds (N = 11) % Seropositive
icterohaemorrhagiae	153	57.1	11	100
bratislava	94	35.1	11	100
autumnalis	9	3.4	6	55
pomona	4	1.5	3	27
hardjo	3	1.1	1	9
copenhageni	2	0.7	2	18
canicola	1	0.4	1	9
javanica	1	0.4	1	9
Any of the above serovars	178	66.4	11	100

Associations between two herd measures of reproductive problems, piglets born dead and nonproductive sow days per parity (NPSD/P) (6), and antibodies to the four most prevalent serovars were evaluated. Two serovars, *L. autumnalis* and *L. pomona*, were evaluated as either present or absent on the farm. The reproductive parameters from these two groups of farms were compared with a *t*-test. Antibodies to the other two serovars, *L. icterohaemorrhagiae* and *L. bratislava*, were found on all farms. Subsequently, least squares regression was used to assess the relationship between the herd prevalence and the level of each reproductive parameter.

There were no significant associations between the percent piglets born dead on a farm and the % hogs with *L. bratislava* titers (r = 0.372, p = 0.324), or the % hogs with *L. icterohaemorrhagiae* titers (r = 0.459, p = 0.214). Farms without *L. pomona* antibody titers averaged 6.0% born dead; this was not significantly different (t = −0.94, p = 0.378) than the average 7.5% born dead on farms with *L. pomona* titers. Farms without *L. autumnalis* titers averaged 5.8% born dead; this was not significantly different (t = −1.02, p = 0.342) than the average 7.4% born dead on farms with *L. autumnalis* titers.

No association was found between the average NPSD/P on a farm and the % hogs with *L. icterohaemorrhagiae* titers (r = 0.431, p = 0.186). Farms without *L. autumnalis* titers averaged 23.1 NPSD/P: this was not significantly different (t = 0.39, p = 0.708) than the average 19.9 NPSD/P on farms with *L. autumnalis* titers. Farms without *L. pomona* antibodies had an average of 14.7 NPSD/P, compared to 39.1 NPSD/P for farms with *L. pomona* titers (t = −5.83, p = 0.0002). Figure 1 shows the relationship between the % hogs with *L. bratislava* titers and NPSD/P (r = 0.738, p = 0.009).

DISCUSSION

The prevalence data in this study were taken from a random sample of herds, although the slaughter hogs were not randomly sampled. Post-exposure titers to leptospirosis persist for more than six months (1), justifying the use of slaughter hogs. Prevalence data can be influenced by vaccination rate, but only one herd had a sporadic history of vaccination, and that herd had vaccinated sows only. Also, vaccine titers are usually less than 1:100 (13,14), and therefore would not be detected by the testing procedure. The leptospiral status of a farm using seroprevalence in slaughter hogs will underestimate the cumulative incidence of leptospirosis in the farm's older sow population (15).

The possibility of cross-reactions obscuring the results in Table I was investigated. The low Kappa (0.23) indicated that there was no substantial difference between the observed agreement between the two serovars, and the agreement expected by chance. This suggests that a cross-reaction between *L. bratislava* and *L. icterohaemorrhagiae* was unlikely. This finding was supported by McNemar's chi-square (p = 0.000), which indicated that there was a significant difference between the prevalence of the two most

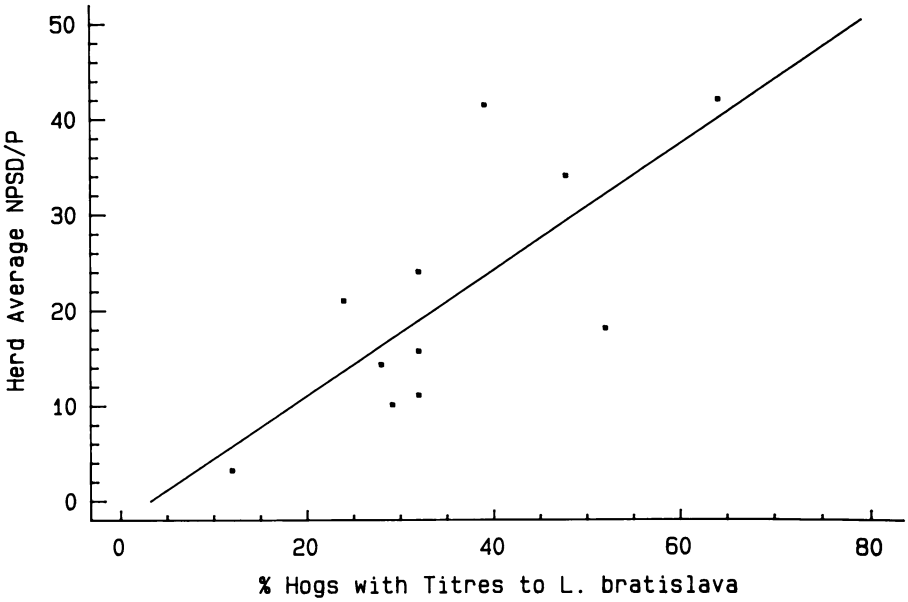

Fig. 1. Association between nonproductive sow days per parity (NPSD/P) and the percent slaughter hogs showing *L. bratislava* antibody titers on 11 Prince Edward Island farrow-finish herds.

common serovars, after controlling for any confounding association between them.

The herd measure of infertility was NPSD/P. Reproductive disease causes abortions, missed heats that lengthen the wean-to-breed interval, reduced farrowing rates and conception rates. All of these increase NPSD/P. Stillbirths, often associated with leptospirosis (1), were measured as % born dead.

For swine herds with clinical signs including abortions, infertility, stillbirths and weak piglets, the most commonly incriminated serotype is *L. pomona* (3,16). The prevalence of *L. pomona* was low in this survey (1.5%, see Table I). This is similar to the prevalence of 1.5% in 138 slaughter sows in Ontario (17). Other prevalences have been reported: 4.6% in 197 Ontario slaughter hogs (2), 0% in 792 hogs in England (3), and 0.7% in 687 hogs from Illinois farms with a history of reproductive problems (4). The risk of stillbirth was no different ($p = 0.378$) on farms with *L. pomona* (7.5% born dead) than on farms without *L. pomona* (6.0% born dead). The lack of significance may be a result of lack of power due to the use of only 11 farms. However, a significant difference ($p = 0.0008$ using the conservative Bonferroni adjustment (10) for multiple comparisons among four serovars) was found between 39.1 NPSD/P on farms with *L. pomona* and 14.7 NPSD/P on farms without *L. pomona*. This suggests that NPSD/P, a measure of infertility, should be considered along with parameters traditionally used to diagnose leptospirosis, such as % born dead and litter scatter.

The most common serovar in this survey was *L. icterohaemorrhagiae* with a prevalence of 57.1%. However, the clinical significance of *L. icterohaemorrhagiae* is dubious. No relationship could be established between the prevalence of reactors on a farm, and the farm's measures of reproduction (% born dead or NPSD/P).

Leptospira bratislava was prevalent in 35.1% of slaughter hogs. This is similar to other prevalences reported: 32% in 197 hogs in Ontario (2), 42% in 762 hogs in Iowa (5), 19.2% in 792 hogs in England (3). The significance of titers to *L. bratislava* is debatable (4,11). On the one hand, this serovar is considered host-adapted to swine, and therefore causes mild infection (18), not associated with a clinical syndrome (19). Titers have been interpreted as the result of cross-reactions with other serovars (11). On the other hand, *L. bratislava* has been isolated from aborted fetuses (20,21), and reproductive disease has resulted from experimental infection (22).

Interpretation of individual titers is difficult; serum antibodies were an unreliable guide to carrier status in cattle (23). Therefore herd level data were used in this study, resulting in farms with higher prevalence of *L. bratislava* titers being associated with infertility, as measured by NPSD/P (see Fig. 1; $r = 0.738$, $p = 0.036$ using the Bonferroni adjustment). These results are similar to an English study that found that *L. australis* (including *bratislava*, *lora* and *australis*) serotiters were associated with infertility in sows (3). There was no association between prevalence of *L. bratislava* titers and % born dead ($p = 0.324$).

Leptospira autumnalis was prevalent in 3.4% of slaughter hogs. The significance of these titers has not been established in other papers. In this study, the lack of a significant association with either % born dead or NPSD/P suggests limited clinical use of *L. autumnalis* titers, although lack of power due to the use of only 11 farms may be a factor.

In summary, both *L. pomona* and *L. bratislava* were associated with infertility, as measured by NPSD/P. Clinical significance could be established for neither *L. icterohaemorrhagiae* nor *L. autumnalis*. None of the four serovars tested was associated with increased stillbirths. This suggests that NPSD/P should be considered along with parameters traditionally used to monitor the effects of leptospirosis (% born dead and litter scatter).

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REFERENCES

1. **HANSON LE, TRIPATHY DN.** Leptospirosis. In: Leman AD, ed. Diseases of Swine. 6th ed. Iowa: Iowa State University Press, 1986: 591-598.
2. **BAKER TF, McEWEN SA, PRESCOTT JF, MEEK AH.** The prevalence of leptospirosis and its association with multifocal interstitial nephritis in swine at slaughter. Can J Vet Res 1989; 53: 290-294.

3. **HATHAWAY SC, LITTLE TWA.** Prevalence and clinical significance of leptospiral antibodies in pigs in England. Vet Rec 1981; 108: 224–228.
4. **HANSON LE.** Bratislava in swine. Proc Am Assoc Swine Pract 1987: 87–91.
5. **HILL H, KINKER D.** Serology interpretation. Proc Am Assoc Swine Pract 1988: 215–222.
6. **VAN TIL LD, DOHOO IR, SPANGLER E, OGILVIE TH.** A survey of biological productivity of Prince Edward Island swine herds. Can J Vet Res 1991; 55: 174–179.
7. **COLE JR, SULZER CR, PURSELL AR.** Improved microtechnique for the leptospiral microscopic agglutination test. Appl Micro Biol 1973; 25: 976–980.
8. **RYAN BF, JOINER BL, RYAN TA.** Minitab Handbook. 2nd ed. Boston: Duxbury Press, 1985.
9. **MARTIN SW, MEEK AH, WILLEBERG P.** Veterinary Epidemiology: Principles and Methods. Ames: Iowa State University Press, 1987.
10. **GLANTZ SA.** Primer of Biostatistics. 2nd ed. New York: McGraw-Hill Book Company, 1987.
11. **HILL H.** Interpretation of serologic results of some important swine diseases. Compend Contin Educ Pract Vet 1988; 10: 979–985.
12. **FLEISS JL.** Statistical Methods for Rates and Proportions. 2nd ed. New York: John Wiley and Sons, 1981.
13. **SANFORDF SE, MORRIS PJ.** Serological titers to various leptospiral serovars before and after vaccinating gilts with three commercial vaccines. Can Vet J 1990; 31: 277–279.
14. **LYNCH J.** Leptospira serology. Ontario Ministry of Agriculture and Food, Veterinary Services Branch: Extension Notes, 1989.
15. **HATHAWAY SC, LITTLE TWA, PRITCHARD DG.** Problems associated with the serological diagnosis of *Leptospira interrogans* serovar *hardjo* infection in bovine populations. Vet Rec 1986; 119: 84–86.
16. **KINGSCOTE BF.** Leptospiral serovars in Canada. Can Vet J 1988; 29: 70–71.
17. **PRESCOTT JF, NICHOLSON VM.** Australis serogroup leptospira in swine abortion. Can Vet J 1987; 28: 312–313.
18. **SONGER JG, THIERMANN AB.** Leptospirosis. J Am Vet Med Assoc 1988; 193: 1250–1254.
19. **ELLIS WA, THIERMANN AB.** Isolation of *Leptospira interrogans* serovar *bratislava* from sows in Iowa. Am J Vet Res 1986; 47: 1458–1460.
20. **ELLIS WA, McPARLAND PJ, BRYSON DG, McNULTY MS.** Leptospires in pig urogenital tracts and fetuses. Vet Rec 1985; 117: 66–67.
21. **ELLIS WA, McPARLAND PJ, BRYSON DG, CASSELLS JA.** Prevalence of *Leptospira* infection in aborted pigs in Northern Ireland. Vet Rec 1986; 118: 63–65.
22. **FARINA R, ANDREANI E, TOLARI F.** Leptospirosis in swine — experimental infection with serotype *bratislava*. Int J Zoonoses 1977; 4: 38–44.
23. **ELLIS WA, SONGER JG, MONTGOMERY J, CASSELS JA.** Prevalence of *Leptospira interrogans* serovar *hardjo* in the genital and urinary tracts of non-pregnant cattle. Vet Rec 1986; 118: 11–13.