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PREVALENCE OF *BRUCELLA* SP. ANTIBODIES IN FERAL SWINE IN FLORIDA

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ABSTRACT: Serum samples collected from feral swine (*Sus scrofa*) throughout Florida (USA) from 1974 to 1989 were tested for antibodies to *Brucella* sp. by the card test, the standard tube test, the rivanol test or the complement fixation test. Seropositive swine were detected at six of 18 sites with a composite prevalence of 23.4% (238 of 1,015 samples; range = 5.5% to 33.3%) for sites with seropositive swine. At one site for which age and sex data were available there was no significant difference ($P = 0.50$) in seroprevalence between males and females. Antibody prevalence in adult (≥8 mo) and juvenile swine (<8 mo), however, was significantly different ($P < 0.05$). Based on these data, *Brucella* sp. infections are limited only to certain populations of feral swine. To avoid the spread of *Brucella* sp. organisms, however, relocation of feral swine is not recommended.

Key words: Feral swine, wild swine, *Sus scrofa*, *Brucella* sp., seroprevalence.

INTRODUCTION

Brucella suis infections have been detected by culture and serology in free-living populations of wild swine in eight states (Arkansas, California, Florida, Georgia, Hawaii, Louisiana, South Carolina, Texas; USA) (Wood et al., 1976; Becker et al., 1978; Clark et al., 1983; Nettles, 1984; Corn et al., 1986; Nettles, 1989). Infections in domestic swine, usually chronic in nature, are characterized by abortion, infertility, orchitis, posterior paralysis and lameness (Deyoe, 1986). The organism is transmitted by oral and venereal routes (Deyoe, 1986).

The State-Federal Cooperative Brucellosis Eradication Program for domestic swine has progressed rapidly. Currently, 38 states are validated brucellosis-free. Between September 1990 and September 1991, 68 infected herds were detected; the majority were located in Alabama, Texas and Florida (U.S. Animal Health Association Committee on Swine Brucellosis, 1991). *Brucella suis* infections in wild swine are important for several reasons. As a potential reservoir of infection, feral swine may seriously jeopardize efforts at eradication from domestic swine. As a zoonotic disease, brucellosis remains a significant threat to humans. *Brucella suis* infections in humans are characterized by fever, chills, headaches and general weakness (Madkour, 1989). The popularity of feral swine as a game animal places the hunter at risk, particularly if adequate protective measures are not followed when field-dressing hogs. Six hunters contracted brucellosis from feral swine in Florida during 1974 and 1975 alone (Bigler et al., 1977). Finally, the incidence of *B. suis* biovar 1 infections in cattle appears to be increasing. During the 5-yr period between 1 October 1982 and 30 September 1987 there were 11 isolations from submissions to the National Veterinary Services Laboratory, Ames, Iowa (USA), as compared to 27 isolations between 1 October 1987 and 30 September 1989 (Payeur et al., 1989). Of these latter 27 isolations, 19 were from Florida cattle.

The distribution, population density and economic value of feral swine in Florida are given by van der Leek et al. (1993). Our objective was to expand the information on the prevalence of *Brucella* sp. antibodies among feral swine in Florida.

MATERIALS AND METHODS

Serum samples were collected from feral swine from 1974 to 1989. A total of 1,327 feral swine from 18 sites were tested (Table 1). Sample collection and preparation are summarized by van der Leek et al. (1992).

Data were available on the sex for 769 of 782 swine sampled in the Fisheating Creek Wildlife Refuge, Glades County, during 1979 and 1980. Data were available on the ages for 313 of 782 swine sampled at the same site. Tooth eruption patterns were used to determine age (Matschke, 1967).

Except for sera collected during 1988, sera were tested by the card test (BBL Microbiology Systems, Cockeysville, Maryland, USA), the standard tube (ST) test, the rivanol test and the complement fixation (CF) test performed as described by Alton (1990). Card tests were interpreted as positive or negative. Standard tube titers ≥1:25, rivanol titers ≥1:25, and CF titers ≥1:20 were regarded as positive. Swine were defined as seropositive if they were positive by at least three of the four tests. Samples collected during 1988 were tested by the card and plate agglutination tests (Alton, 1990) only, due to limited availability of serum. Plate test results were interpreted as positive or negative at a 1:25 dilution. Swine collected in 1988 were defined as seropositive if they were positive by both of these latter two tests. However, 24 of 132 samples produced card test results that could not be interpreted due to hemolysis. Samples which were positive by the plate test, but which could not be examined by the card test were interpreted as suspect.

The Chi-square test was used to evaluate differences in prevalence between males and females, and between adults and juveniles, using a proprietary computer software program (EPISTAT©, Round Rock, Texas). The significance level was set at $\alpha = 0.05$.

RESULTS

Swine with antibodies to *Brucella* sp. were detected at six (33%) of 18 sites (Table 1, Fig. 1). The composite prevalence was 23.4% (238 of 1,015 samples; range = 5.5% to 33.3%) for the six sites with seropositive swine. Suspect swine were detected at Tosohatchee WMA, Orange County ($n = 1$), and Cecil M. Webb WMA, Charlotte County ($n = 3$), during 1988.

Prevalence of antibodies in total males (28.0%) versus total females (25.8%), adult males (49.3%) versus adult females (37.5%), and juvenile males (17.6%) versus juvenile females (7.6%) were not significantly different ($P = 0.54$, 0.24 and 0.70, respectively) in swine from the Fisheating Creek Wildlife Refuge, Glades County. Antibody prevalence in adults (44.3%) was significantly higher than in juveniles (12.6%) ($P < 0.05$).

Comparing the 1979 and 1980 Fisheating Creek Wildlife Refuge samples by individual test or combination of tests, the seroprevalence as determined by the card test alone (26.2%) approximated the seroprevalence as determined using three separate tests (27.6%) (Table 2).

DISCUSSION

Definitive evidence of brucellosis is obtained only by isolation and identification of *Brucella* sp. Since this is not always possible, serological tests provide supporting evidence in the diagnosis of brucellosis.

Free-living feral swine should be considered as originating from a herd of unknown status and therefore swine with ST test titers ≥1:25 should be classified positive (U.S. Department of Agriculture, 1986). Due to the occurrence of nonspecific (heterospecific) reactions at this dilution (Alton, 1990), other workers have used a battery of tests to detect *Brucella*-infected wild swine. Our results (Table 2) were consistent with the heterospecific phenomenon described by Alton (1990) with the ST test having the highest seroprevalence when used alone. When sensitivity is increased by the addition of other tests, the prevalence decreases. Cumulatively, our data (Table 2), combined with published data as presented below, support the idea that the card test alone gives a suitable estimate of the number of *Brucella* sp. infected wild swine at a particular site on an individual basis; however, the

TABLE 1. Prevalence of *Brucella* sp. antibodies in feral swine in Florida.

Counties[a]	Site	Years sampled	Tests used	Number tested	Number positive	Prevalence (%)
1) Franklin	St. Vincent's Isle (29°40′N, 84°38′W)	1974, 1976, 1978	Card, ST[b], CF[c], rivanol	10	0	
2) Taylor, Wakulla, Jefferson	Aucilla WMA[d] (30°10′N, 84°04′W)	1988	Card, plate	17	0	
3) Taylor	Tide Swamp WMA (29°50′N, 83°30′W)	1988	Card, plate	2	0	
4) Columbia	O'Leno State Park (29°50′N, 82°37′W)	1977, 1979	Card, ST, CF, rivanol	17	0	
5) St. Johns	Guana River WMA (30°05′N, 81°20′W)	1988	Card, plate	4	0	
6) Dixie, Lafayette	Steinhatchee WMA (29°50′N, 83°15′W)	1978	Card, ST, CF, rivanol	7	1	14
7) Alachua	Orange Heights (29°45′N, 82°06′W)	1979	Card, ST, CF, rivanol	11	0	
8) Levy	Brunswick (29°17′N, 83°05′W)	1977 to 1979	Card, ST, CF, rivanol	10	3	30
9) Polk, Sumter, Lake	Green Swamp WMA (28°20′N, 81°00′W)	1988	Card, plate	9	3	33
10) Orange	Tosohatchee WMA (28°30′N, 81°00′W)	1979, 1980[e]	Card, ST, plate, CF, rivanol	10	2	20
		1988	Card, plate	9	0	
11) Osceola	Bull Creek WMA (27°55′N, 81°00′W)	1988	Card, plate	12	0	
12) Orange, Osceola, Brevard	Deseret Ranch (28°15′N, 81°00′W)	1979, 1980[e]	Card, ST, plate, CF, rivanol	10	3	30
13) Osceola	Prairie Lakes State Park/Three Lakes WMA[f] (28°00′N, 81°15′W)	1979, 1980[e]	Card, ST, plate, CF, rivanol	10	3	30
		1988	Card, plate	3	0	
14) Hardee	Ona (27°28′N, 81°53′W)	1977	Card, ST, CF, rivanol	2	0	
15) Sarasota	Myakka River State Park (27°15′N, 82°15′W)	1979, 1980[e]	Card, ST, plate, CF, rivanol	24	0	
		1989	Card, ST, plate, rivanol	43	6	14
16) De Soto	Bright Hour Ranch (27°05′N, 81°40′W)	1978	Card, ST, CF, rivanol	164	9	6
17) Charlotte	Cecil M. Webb WMA (26°50′N, 81°55′W)	1988	Card, plate	58	0	
18) Glades	Fisheating Creek Wildlife Refuge (26°50′N, 81°55′W)	1977, 1978[g]	Card, ST, CF, rivanol	95	26	27
		1979, 1980	Card, ST, CF, rivanol	782	216	28
19) Palm Beach	J.W. Corbett WMA (26°50′N, 81°10′W)	1988	Card, plate	18	0	

[a] Counties are listed north to south starting in the Florida Panhandle.
[b] ST, standard tube test.
[c] CF, complement fixation test.
[d] WMA, Wildlife management area.
[e] Data collected by Zygmont et al., 1982; samples listed as originating from Orange County originated from a pooled Orange/Brevard County shipment.
[f] Sites are immediately adjacent and considered one site.
[g] Data collected by Becker et al., 1978.

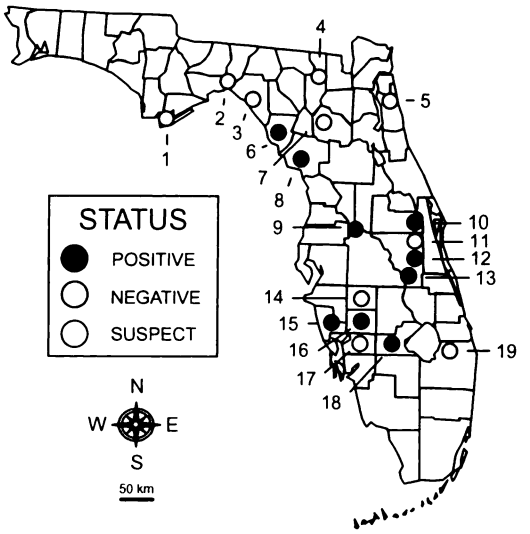

FIGURE 1. Distribution of sites with feral swine in Florida positive for *Brucella* sp. antibodies (numbers are keyed to Table 1; site number 10 also had suspect swine). Includes data from Becker et al. (1978) and Zygmont et al. (1982).

status of a wild hog would depend on the test used. The card test avoids the heterospecificity encountered when using the ST or plate tests and the anticomplementary activity sometimes encountered on the CF test. More importantly, the card test is easy to perform and can be used in the field.

In South Carolina, Wood et al. (1976) used the CF, rivanol and card tests. Of 255 swine they tested, 18.0% were positive by the card test alone. The card test results differed from other test results for only three sera. In Florida, Becker et al. (1978) used the ST test (1:25 dilution), CF test (1:20 dilution), card test and rivanol (1:25 dilution) test. Of 95 swine they tested, 52.6% were seropositive by at least one test, 27.4% were positive by at least one of three tests and 19.0% were positive by the card test alone. In Texas, Corn et al. (1986) used the ST (1:100 dilution), CF (1:20 dilution), card, plate (1:100 dilution), buffered plate (1:25 dilution) and rivanol (1:25 dilution) tests. Of 124 swine they tested, 13.7% were seropositive by at least one test, 5.6% were positive by at least three tests and 4.8% were positive by the card test alone. Testing swine from Hawaii and several southeastern states, Zygmont et al. (1982) used the same six tests. Of 352 swine they tested, 10.2% were positive by at least one test, 6.0% were positive by at least three tests and 7.1% were positive by the card test alone.

TABLE 2. Serologic test results of 782 feral swine from Fisheating Creek Wildlife Refuge, Glades County.

Test or combination of tests	Number positive	Percent positive
Card only	205	26.2
Standard tube only	391	50.0
Complement fixation only	287	36.7
Rivanol only	224	28.6
Any one test	448	57.3
Any two tests	293	37.5
Any three tests	216	27.6
All four tests	150	19.2

When the data of Becker et al. (1978) and Zygmont et al. (1982) are included, swine with *Brucella* sp. antibodies were detected at 9 of 19 sites between 1974 and 1989, with a composite prevalence of 23.2% (Table 1, Fig. 1). The sites with seropositive feral swine are spread throughout the state with two of these sites located near to the predominant region of domestic swine production in north central Florida.

Two sites (Tosohatchee WMA, Orange County; and Prairie Lakes State Park/Three Lakes WMA, Osceola County) previously identified as containing seropositive pigs (Zygmont et al., 1982) did not contain any seropositive pigs during the 1988 sampling, although one suspect sample originated from the Tosohatchee WMA. Conversely, one site (Myakka River State Park, Sarasota County) previously identified as containing no seropositive pigs (Zygmont et al., 1982) had seropositive pigs during the 1989 sampling. This may reflect the introduction of *Brucella* sp. since the earlier sampling. However, the data must be interpreted in light of the sample size per site and the criteria used to define seropositive swine. Several sites with small

sample sizes had no seropositive swine, but further testing is needed to confirm the absence of brucellosis at these sites.

In earlier studies there was a significantly higher seroprevalence of *Brucella* sp. antibodies in adult swine compared to juvenile swine, but no difference in seroprevalence reported between males and females of all ages (Wood et al., 1976; Becker et al., 1978). In our study, more juvenile males (17.6%) were seropositive than juvenile females (7.6%), although this was not significant, most likely due to the small sample size.

The presence of brucellosis in Florida feral swine is significant for several reasons. The relocation of feral swine within the state may result in the transmission of infection to naive wild swine populations. The introduction of feral swine into backyard domestic swine herds, as commonly occurs in the southeastern USA, may result in the introduction of brucellosis and could seriously undermine the State-Federal Cooperative Eradication Program. Although many feral swine sites did not contain swine with *Brucella* sp. antibodies, hunters are urged to use caution when handling feral swine carcasses to minimize the risk of contracting this serious zoonotic disease. Finally, as for pseudorabies virus, the brucellosis status of a population of feral swine would best be determined by testing adult swine.

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