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THE OCCURRENCE OF BRUCELLA AGGLUTININS IN CATTLE IN THE PANAMA CANAL ZONE*

By EDWIN O. JORDAN *and* J. MCBROOM
Gorgas Memorial Laboratory, Panama, Canal Zone

A collection of serum from the Mindi Dairy herd, in the Panama Canal Zone, in February, 1931, afforded an opportunity for determining the presence of Brucella agglutinins in cattle in this region.† The herd consisted of purebred and grade Holstein and Jersey cattle. The majority of these animals were born on the Isthmus; those brought in from the United States (mostly purebred Holsteins) were from 2 to 6 years old. The herd was under constant and expert veterinary care. The serum was drawn with antiseptic precautions and kept at ice-box temperature except for the few days it was in transit to the laboratory. Phenol (0.5 per cent) was added as a preservative. The usual macroscopic method was used; the mixture of saline suspension and serum was incubated at 37° C. for 2 hours and then kept at ice-box temperature overnight before the readings were made. The period of testing was unavoidably extended over several months, but a number of sera were tested at the beginning of the period and then retested later; the results show that in some cases a slight loss of agglutination strength occurred but that the general tabulations were not affected. Six Brucella strains were used in preliminary tests. One of them, of porcine origin, proved so sensitive that it agglutinated to some degree with all sera tested; another, a human strain, also showed a tendency to agglutinate in relatively high dilutions. As other observers have found, the selection of suitable antigens is a matter of primary importance. Two strains, one from the liver of an aborted pig, obtained from Dr. J. W. Connaway, University of Missouri, and the other from an aborted bovine fetus, obtained from the U. S. Bureau of Animal Industry, Washington, D. C., were finally isolated and gave uniform and consistent results throughout. Rarely one of these strains would be agglutinated at a 1:40 dilution while the other was not, but in almost all instances the readings were identical. Living antigens were used in all tests, partly for the purpose of being able to correlate the results with those of another serological study

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which was in progress in which this procedure seemed desirable. The turbidity of the antigen suspension was kept uniform throughout by comparison with a barium sulfate suspension. Platings were made at frequent intervals to insure smoothness. Comparative tests made with a standard Brucella antigen, obtained from Dr. K. F. Meyer, showed no essential differences in the agglutination titres. The sera of 383 cows were examined, with the results shown in table I.

TABLE I.—*Summary of agglutination tests.*

Agglutination Reaction	Animals	
	No.	%
Positive in dilution of 1:100 or higher	52	13.6
Positive in dilution of 1:80, but not at 1:100	43	11.2
Negative in dilution of 1:80	288	75.2
Totals	383	100.0

These results are quite similar to those obtained in other localities, but show a somewhat lower proportion of positive reactions than those recorded, for example, by Norten and Pless,[1] in Michigan (21 per cent positive), or by Hardy *et al.*,[2] in Iowa (26 per cent positive, i. e., 1:80 or higher). The exact comparative results are shown in table II.

TABLE II.—*Results obtained in three different localities.*

Locality	Cows Tested	Positive						Negative	
		High Dilution			Low Dilution				
		No.	%	Titre	No.	%	Titre	No.	%
Panama Canal Zone	383	52	13.6	1:100+	43	11.2	1:40	288	75.2
Michigan	647	136	21.0	1:100+	115	17.8	1:25 1:50	396	61.2
Iowa	1389	339	26.0	1:80 +	105	8.0	1:40	856	66.0

If a positive agglutination reaction in a dilution of 1:100 or higher is taken as evidence of infection with Brucella, and absence of reaction in a dilution of 1:40 as evidence of freedom from infection, then the dairy cows in the Canal Zone appear to have been somewhat less generally infected at the time of this test (February, 1931) than those in some other localities.

Enough serum was available for determining the upper titre limit of the sera in 46 of the 52 cases in which agglutination occurred at 1:100 and over. The results are given in table III.

TABLE III—*Upper titre limits of 46 reactors.*

UPPER TITRE LIMIT	REACTORS
1:100	19
1:200	10
1:400	6
1:800	8
1:1600	1
1:3200	2
Total	46

Twenty-two of these 46 sera gave a very definite zoning effect and a few others gave indications of zoning with one or more strains. This was often very marked, as is shown by the examples in table IV. Several zoning sera were treated with the prepared antigen sent us by Dr. Meyer and gave similar results.

TABLE IV—*Zoning effect shown by four sera.*

SERUM	DILUTION					
	1:40	1:100	1:200	1:400	1:800	1:1600
170	−	+	++	tr	.....	.....
133	−	++	+++	+++	tr	−
135	−	tr	+	+++	+	−
141*	−	−	++	++	.....	.....

*In nearly all instances where zoning was observed, the dilution 1:100 showed at least a trace of agglutination. Serum 144 is an exception.

Exact histories with respect to abortion were available for 329 of the 383 cows. Sixty-nine of these had records of from one to three abortions; 260 had no history of abortion. Of the 69 aborting animals, 37.7 per cent gave a serum reacting in a dilution of 1:40 or higher (10.1 per cent, 1:40; 27.5 per cent, 1:100 or higher). Of those with no history of abortion, 21.5 per cent gave a serum reacting at 1:40 or higher (8.8 per cent, 1:40; 12.7 per cent, 1:100 or higher). The occurrence of agglutinins in the serum of cows that have never aborted has been noted by Norton and Pless,[1] Huddleson and Smith,[2] and others. The sera of 14 bulls were tested: all were negative in a 1:40 dilution. The sera of 12 yearling heifers were tested and proved

negative in a 1:50 dilution in 10 cases and positive at 1:40 (and not higher) in 2.

The sera of 28 men, engaged in handling milk and caring for the animals, etc., were tested; all but one were negative in a 1:40 dilution. The single positive reactor (D. G.) was a man about 62 years old who had been a cattleman all his life and was at the time employed in handling the breeding herd and calves. He gave no definite history of undulant fever but had been in the Ancon Hospital, with diagnoses, on separate occasions, of malaria complicated with bronchitis, and amoebic dysentery. No case of undulant fever, traceable to cattle, seems yet to have been recorded in the Zone.

SUMMARY

Brucella abortus infection, as determined by the agglutination test, is present in dairy cattle in the Panama Canal Zone, although apparently to a somewhat less degree than in many parts of the United States. Reactions at 1:100 and higher are more than twice as common in animals that have aborted as in those with no history of abortion. Undulant fever in man, traceable to cattle, has not yet been recorded in the Zone.

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[3]Huddleson, I. F., and Smith, L. H.: Jour. A. V. M. A., lxxix (1931), n.s. 32 (5), pp. 63-78.