

BODY OF REPORT

SEATO Medic Study No. 80C Isolation of Leptospires from Thailand Domestic Livestock Abattoir Study Utilizing Direct Kidney Culture.

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FINAL REPORT

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Objective: To examine by direct culture kidneys from cattle, buffalo and swine presented to the Bangkok Abattoir from provinces in Thailand. Correlation of isolates obtained with serological agglutinin response is accomplished.

Description: Swine, cattle and buffalo are presented to the Bangkok Abattoir from many of the provinces of Thailand. The market age for swine is one year; for cattle and buffalo 6 years or older. The younger age of swine should be better for leptospira recovery. To attempt to isolate some of the organisms which are eliciting a very diversified agglutinin response in domestic livestock of Thailand, a kidney sampling study was started at the Bangkok Abattoir. Animals are selected prior to slaughter according to province of origin. An attempt is made to select animals from various provinces. Blood for serum is collected at the exsanguination station. At this point a tag is placed on the animal giving province of origin and an estimate of age. When the tagged animals reach the point where the body wall is opened, one kidney is removed aseptically and placed in a sterile specimen box. The kidneys are transported to the laboratory (about 5 km. distance) and placed in the refrigerator until processed. Only 1 1/2 to 3 1/2 hours elapse before the last kidney is cultured. In the laboratory the perirenal fat is removed and discarded. The kidney is halved mid-sagittally and placed with both cortical surfaces up. The cortical surfaces are flamed with a bunsen burner until the surface turns tan colored. Using a new scalpel, the discolored surface layer of the cortex is removed and special constructed kidney scrapers are used to scrape the remaining cortical surface. Collected within the scraper is a musky cortical homogenate. One drop of a 1:100 and 1:1000 dilution of this homogenate in liquid Stuart's media is inoculated into three (3) tubes each of semi-solid Fletcher's media containing eight (8) , twelve (12) and sixteen (16) per cent rabbit serum and ten (10) per cent agglutinin free homologous species serum. The cultures are examined by darkfield microscopy at fifteen (15) and thirty (30) days for the presence of leptospire.

Results: One isolate was obtained from three hundred and seventy-nine (379) buffalo and three (3) isolated were obtained from four hundred and twenty eight (428) cattle kidneys examined from thirty-five (35) provinces respectively. From three hundred and twelve (312) swine kidneys, twenty four (24) isolates were obtained. There were twenty-two (22) isolates from one hundred and seventy-three (173) swine (12.6% recovery rate) from eight (8) provinces of the central plain area two (2) isolates from one hundred and thirty-nine (139) swine (1.4% recovery rate) from seven (7) provinces beyond the central plain area. The isolates are summarized in Table I.

Recovery rate was equally good at the 1:100 and 1:1000 dilution of kidney homogenate. The eight (8) per cent and twelve (12) per cent rabbit serum gave about the same recovery rate, but the sixteen (16) per cent rabbit serum was less efficient. The homologous serum was inferior to rabbit serum (two (2) of twenty four (24) swine isolates and no cattle or buffalo isolated recovered).

The low isolate recovery rate encountered in attempting to isolate leptospire

Table I
SUMMARY OF ISOLATE RESULTS

Species	# Isolates	Serotype	Homologous Serum Reaction
Swine	15	po $mona$	Negative *
	3	po $mona$	autumnalis 1:10
	2	po $mona$	po $mona$ 1:10
	1	po $mona$	bataviae 1:10
	1	po $mona$	borincana 1:10
	1	canicola	negative *
	1	javanica	negative *
Buffalo	1	alex i	grippotyphosa 1:25
			autumnalis 1:25
			alex i 1:25
Cattle	1	alex i	alex i 1:100
	1	autumnalis	andaman 1:25
	1	autumnalis	negative * autumnalis 1:25

* Negative to all serotypes

from freshly collected buffalo and cattle kidneys in contrast to the higher recovery rates from swine kidneys using the same techniques is not easily explained. Since by serology the per cent of reactors is higher in buffalo and cattle than in swine, it is unlikely that the infection rate would be higher in swine than in cattle and buffalo. It is possible that cattle and buffalo, although carrying fewer active infections at time of slaughter have previously been exposed to a greater number of leptospiral serotypes, whereas the swine slaughtered at one year of age, while having more active infections at time of slaughter, have had a shorter time to develop leptospiral agglutinins. It is also possible that the swine may simply not respond as well immunologically to leptospira infection and therefore have a greater incidence of infection than determined serologically as well as being more subject to persistent infection. A third possibility may be a variation of the strains isolated. Most of the swine isolates were L. pomona which is known to be well adapted to the swine kidney and can be shed for long periods of time, whereas the isolates L. alexi and L. autumnalis obtained from cattle and buffalo may only produce kidney infections of short duration. Overall, the results from this study, in the light of the much higher reaction rate and varied response of serologic studies, were disappointing. The correlation of isolates to serologic reactions cannot account for the very varied serologic response obtained.

Summary: Kidneys from three hundred and twelve (312) swine, three hundred and seventy-nine (379) buffalo and four hundred and twenty-eight (428) cattle have been examined for the presence of the leptospire organism by direct kidney culture techniques. Isolates were recovered from twenty-four (24) swine, one (1) buffalo and three (3) cattle kidneys. These isolates were predominantly L. pomona in swine and L. alexi and L. autumnalis in cattle and buffalo. This is a final report.