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Source: Journal of Wildlife Diseases, 15(4) : 529-532

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-15.4.529

YERSINIAE IN THE SOIL OF AN INFECTED WAPITI RANGE

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Abstract: Yersinia enterocolitica was isolated from 10 of 121 soil samples from an area inhabited by infected wapiti *(Cervus elaphus roosevelti)* in northwest California. Significantly ($p<0.05$) more soil samples from a forest habitat were infected, compared to soil samples from prairie habitats. Soil was found infected with yersiniae only on dates for which rainfall in excess of 17 mm had occurred during the previous 7 days.

INTRODUCTION

Yersiniosis is a wildlife mortality factor receiving increased attention from biologists. Although animals have been viewed as the reservoir of *Yersinia pseudotuberculosis*,[7] it has been suggested that soil or water may be a reservoir of *Yersinia enterocolitica*.[2,8] Recently four strains were isolated from soil and leaves in France.[1]

Yersinia enterocolitica was isolated from the feces of apparently healthy Roosevelt wapiti *(Cervus elaphus roosevelti)* at Prairie Creek State Park, Humboldt County, California.[8] In the present study, the isolation of yersiniae directly from soil of the sites inhabited by infected wapiti is reported.

MATERIALS AND METHODS

Boyes Prairie and Gold Bluffs Beach are two areas used by wapiti known to be infected with *Y. enterocolitica*. These sites have been described previously.[4,5,6] Three to five soil samples were taken from a one hectare area at the north end of Boyes Prairie on each of 10 different weeks between 29 March and 21 June 1977. Two samples were taken concurrently from each of four sites on Gold Bluffs Beach: a low-lying wet meadow, a moderately wet grassy site and a relatively dry sandy hillock on the coastal prairie,[9] as well as an alder thicket in the coastal forest;[5,9] these sites were adjacent to the coastal strand.[9]

For each sample, a 2 to 3 g plug of surface soil, along with any accompanying vegetation, was collected and stored at 4 C for 9 to 10 months in 9 ml trypticase soy broth with 0.5% yeast extract. Care was taken to avoid taking soil samples with any evidence of feces. MacConkey agar and trypticase soy agar were used for initial isolations. Suspect colonies were characterized by standard materials and methods.[3] The methyl red and acetylmethylcarbinol tests were done at both 20 C for 5 days and 37 C for 2 days. One percent concentrations of filter-sterilized carbohydrates in purple broth base were used, except that 1% concentrations of glucose and lactose in the O-F tests and a 0.5% concentration of soluble starch were autoclaved for 10 minutes at 121 C. A selection of the organisms was sent to the Public Health Service Laboratories, Fort Collins, Colorado and evaluated with antisera to *Y. enterocolitica* serotypes 1 to 21, 24, 32, the Arizona and Tacoma types, as well as to *Y. pseudotuberculosis* serotypes I through VI.

RESULTS

Yersinia enterocolitica was isolated from 10 (8.3%) of 121 soil samples taken. All of the isolates were gram-negative rods motile at 20 C and non-motile at 37 C. All were positive for urease, catalase, ONPG, and ornithine decarboxylase; all were + or ± for the methyl red test at 37 C.

TABLE 1. Variation among the *Yersinia enterocolitica* isolated in 1977 from soil at Prairie Creek State Park, Humboldt County, California.

	Alder Thicket							Meadow		Grassy Area		Boyes Prairie	
Characteristics	A1		A2	A6	A12		A17	M1	M17	G12	G18*	P2	
Date Collected	29 Mar		29 Mar	12 Apr	3 May		31 May	29 Mar	31 May	3 May	31 May	29 Mar	
Somatic Serotype	5	ns	17	4,32	5		4,32	5	5	20	ns	ns	ns
Methyl red (20°C)	−	+	−	V	−	+	+	+	−	+	+	+	−
Acetylmethyl- (20°C)	+	+	V	+	+	−	−	V	+	−	−	−	+
carbinol (37°C)	−	+	−	−	−	−	−	V	−	−	−	−	−
Indole	+	+	+	+	+	−	−	+	V	−	−	−	+
Simmon's Citrate	−	−	+	−	−	−	−	−	−	−	−	−	−
Lactose 0-F	0	0	0	0	0	0	0	0	0	0	−	0	0
Carbohydrate Fermentation (14 days)													
Rhamnose	−	−	+	−	−	−	−	−	−	−	−	+	+
Sucrose	+	+	+	+	+	+	+	+	+	+	+	−	+
Melezitose	−	+	−	−	−	−	−	−	−	−	−	−	−
Raffinose	−	+	−	−	V	−	−	−	−	−	+	−	−
Inulin	−	+	−	−	−	−	−	−	−	−	−	−	−
Soluble starch	V	+	+	+	+	+	+	+	V	+	+	+	+
Salicin	+	+	+	+	+	−	V	+	+	V	+	+	+
Inositol	+	+	V	+	+	−	−	V	V	−	−	V	+

* Sample contaminated with Boyes Prairie soil

+: positive reaction
−: negative reaction
V: reaction varies for individual isolates
ns: not serotypable with the *Yersinia enterocolitica* or *Y. pseudotuberculosis* antisera used
O: oxidative

All were negative for H_2S in KIA, oxidase, gelatinase, phynylalanine deaminase, lysine decarboxylase and arginine dihydrolase; none utilized malonate. They all fermented glucose in the O-F test. All isolates utilized arabinose, fructose, galactose, cellobiose, trehalose, sorbitol and mannitol in 48 h; all used xylose and glycerol in 7 days. None used dulcitol in 14 days. Reactions varied with the other tests (Table 1).

The yersiniae were found in 5 of 20 samples from the alder thicket, 2 of 20 from the meadow community, one of 18 from the grassy area, none of 18 from the hillock and one of 45 samples from Boyes Prairie. An additional strain was isolated from a grassy area soil sample contaminated with soil from Boyes Prairie. Based on an adjusted G test,[9] the frequency of *Y. enterocolitica* isolation from the alder thicket was significantly ($p<0.05$) higher compared to the frequency of isolation from the other sites, collectively. This suggests an association of the yersiniae with the coastal forest, rather than with a prairie ecosystem.

The isolation frequency of the yersiniae was also related to rainfall. Weekly rainfall varied from one to 53 mm during the period of the study. During each of the four dates (Table 1) that yersiniae were isolated, rainfall in excess of 17 mm fell during the previous 7 days. But for the six dates that yersiniae were not isolated (5, 19, 26 April; 10, 17 May; 21 June), rainfall in excess of 9 mm occurred only once during the previous 7 days. Based on an adjusted G test,[10] this difference was significant ($p<0.05$). Previous isolations of yersiniae from Prairie Creek wapiti[8] also were made following a period of higher rainfall.

DISCUSSION

The isolation of *Y. enterocolitica* strains from soil in this study that appear to be biochemically and serologically identical to strains isolated from wapiti on the sample sites[8] points to the likelihood of *Y. enterocolitica* transmission between soil and wapiti. It is not clear from these data whether soil or wapiti are the original source. But, there is some information to suggest that the wapiti may not have been an important source of the yersiniae in the soil.

The occurrence of yersiniae in soil of an area was not correlated to the observed daytime use by wapiti or to the frequency of infected wapiti feces on that area. For example, the alder thicket, which had the highest frequency of soil yersiniae, is an area used infrequently by wapiti throughout the year.[4,5] In contrast, the Boyes Prairie study area has a heavy use by wapiti,[6] and the wapiti on the area previously were found to have a high frequency of yersiniae (7 of 16 fecal samples[8]); yet only one of 45 soil samples from this site had detectable yersiniae.

In this study, *Y. enterocolitica* was consistently isolated during the spring from soil of a site on the coastal forest. One way to establish the hypothesis that soil is a reservoir for yersiniae is to demonstrate that the bacteria can be found in soil of a site throughout the year, and can multiply in soil at a rate adequate to replace annual attrition.

Acknowledgement

I appreciate the aid of Dr. Thomas J. Quan in evaluating the serotypes of the isolates and critically reviewing a draft of this manuscript.

LITERATURE CITED

1. BARRE, N., C. LOUZIS, M. TREIGNIER, J.M. ALONSO, H. BERCOVIER and H.H. MOLLARET. 1976. Premiers isolements de *Yersinia enterocolitica* a partir d'échantillos de sols. Med. Maladies Infect. 6: 520-521.

2. BOTZLER, R.G., T.F. WETZLER, A.B. COWAN and T.J. QUAN. 1976. Yersiniae in pond water and snails. J. Wildl. Dis. 12: 492-496.
3. COWAN, S.T. and K.J. STEEL. 1965. *Manual for the Identification of Medical Bacteria*. Cambridge University Press, London. 217 p.
4. FRANKLIN, W.L. 1968. Herd organization, territoriality, movements and home range in Roosevelt elk. M.S. Thesis, Humboldt State College, Arcata, Calif. 89 p.
5. ——, A.S. MOSSMAN and M. DOLE. 1975. Social organization and home range of Roosevelt elk. J. Mammal. 56: 102-118.
6. HARPER, J.A., J.H. HARN, W.W. BENTLEY and C.F. YOCOM. 1967. The status and ecology of Roosevelt elk in California. Wildl. Monogr. No. 16.
7. MAIR, N.S. 1973. Yersiniosis in wildlife and its public health implications. J. Wildl. Dis. 9: 64-71.
8. MARTYNY, J.W. and R.G. BOTZLER. 1976. Yersiniae isolated from wapiti *(Cervus canadensis roosevelti)*. J. Wildl. Dis. 12: 386-389.
9. MUNZ, P.A. 1963. *A California Flora*. Univ. Calif. Press, Berkeley.
10. SOKAL, R.R. and F.J. ROHLF. 1969. *Biometry*. W.H. Freeman and Co., San Francisco. 176 p.

Received for publication 11 September 1978