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## *Yersinia enterocolitica* AND *Yersinia pseudotuberculosis* FROM WILDLIFE IN ONTARIO<sup>1</sup>

M. A. HACKING and L. SILEO<sup>2</sup>

**Abstract:** Four isolates of *Yersinia enterocolitica* and six of *Y. pseudotuberculosis* were made from carcasses collected in Ontario in 1973. *Y. enterocolitica* was isolated from the following species: Canada goose, *Branta canadensis* (serotype 4,33), Pekin robin, *Leiothrix lutea* (serotype 6,30), beaver, *Castor canadensis* (serotype as yet unknown), and raccoon, *Procyon lotor* (serotype 5,27). Yersiniosis was apparently the cause of death of the beaver and the cause of liver abscesses in the raccoon; the significance of the isolates from the birds was not determined. *Y. pseudotuberculosis* isolations were from a crow, *Corvus brachyrhynchos* (serotype 1A), two purple martins, *Progne subis* (serotype 1B), and three beavers (serotype 1B). Yersiniosis was apparently the cause of abscesses in one of the beavers and the cause of death of the other cases.

During the period from 1962 to 1972, *Y. pseudotuberculosis* had been isolated in this laboratory from four beavers and one snowshoe hare, *Lepus americanus*. There was no record of previous isolation of *Y. enterocolitica*. In most cases, septic abscesses were found in various tissues; however, significant pathological findings were absent in the Canada goose and the Pekin robin. All but the Pekin robin were free-living native animals.

### INTRODUCTION

The genus *Yersinia* (formerly included in the *Pasteurella*) is placed in the family Enterobacteriaceae in the 8th edition of Bergey's Manual of Determinative Bacteriology. It contains three species, *Y. pestis*, *Y. enterocolitica*, and *Y. pseudotuberculosis*. Yersiniosis refers to infection with the latter two.

Since its recognition as a distinct species in the 1960's, *Y. enterocolitica* has been isolated from many sources, including apparently normal animals.<sup>4,20</sup> It has been isolated from warm-blooded and cold-blooded vertebrates, as summarized by Langford,<sup>12</sup> invertebrates,<sup>16</sup> and from water.<sup>18,16</sup> Its role as a human pathogen has been increasingly recognized in recent years.<sup>3</sup> There are few reports of *Y. enterocolitica* from free-living vertebrate wildlife. Niléhn,<sup>15</sup> in Europe, stu-

died 23 strains from hares and one from a rabbit. Dubois *et al.*<sup>6</sup> isolated this organism from 14 of 100 hares surveyed in Belgium. Three strains were found by Wetzler and Hubbert<sup>20</sup> in feces from apparently normal deer in Michigan, and one strain by Botzler *et al.*<sup>4</sup> from a leopard frog in the same area. There are even fewer reports from birds. Halen and Meulemans<sup>5</sup> reported isolations from a goldfinch and a canary in the same aviary in Belgium, and Langford<sup>12</sup> reported isolations from two pigeons in British Columbia.

*Y. pseudotuberculosis* has also been reported from many species. In Britain, where Mair reported isolations from 21 species of wild birds, it is considered of greater importance in birds than in mammals.<sup>14</sup> This has not been the case in North America. In his review, Hub-

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bert mentions three reports from wild birds: an unknown species of blackbird in 1940, purple grackles in 1962, and a wild pigeon in 1965.<sup>9</sup> Isolations from wild mammals have included the black-tailed jack rabbit, white-tailed antelope squirrel, eastern cottontail, beaver, muskrat, raccoon, mule deer, Norway rat, and snowshoe hare.<sup>10,11</sup>

Four isolations of *Y. enterocolitica* and six of *Y. pseudotuberculosis* were made from carcasses collected in Ontario in 1973 and submitted for diagnosis.

#### MATERIALS AND METHODS

Necropsies were performed on all the carcasses. In those carcasses suitable for histological examination, selected tissues were fixed in 10% buffered formalin, processed by standard paraffin embedding methods, and sections were stained with hematoxylin and eosin. Bacteriological cultures were inoculated from various tissues using blood agar and MacConkey agar. When pathogenic bacteria were not found on initial culture but lesions suggested an infectious process, saline suspensions of liver tissue were inoculated intraperitoneally into guinea pigs. *Yersinia* isolates were sent to the Laboratory Services Branch, Ontario Ministry of Health, Toronto, for identification and serotyping.

#### CASES

##### *Y. pseudotuberculosis*

**695-73:** One immature male crow was found in Bruce county unable to fly. The crow was held in captivity for 3 days, became increasingly lethargic, and died on July 19. At necropsy, nodules of white caseous material 1-5mm in diameter were noted in the skin, pectoral muscle, liver, spleen, lungs, and humeroulnar joint. There were white, caseous plaques on the oral and intestinal mucosa and anterior airsacs. Histological examination revealed ulceration of epithelial tissue and abscessation of other tissues. Numerous bacterial colonies

were associated with these lesions. An interesting finding in one skin nodule was the presence of large eosinophilic intracytoplasmic inclusion bodies associated with marked epidermal hyperplasia. This lesion was the basis of a diagnosis of avian pox. *Y. pseudotuberculosis* type 1A was isolated from lesions in the liver, spleen, lung, and intertarsal joint.

**848-73 and 849-73:** An entire colony of purple martins, consisting of approximately 50 nesting pairs, was lost over a 6 week period in Kent county. The owner of the nest structure collected approximately 30 dead and dying birds in the first week of July when the epizootic began. Two birds were submitted frozen, an immature male and an adult female. Both spleens were enlarged (4 x 15mm in one); both livers and spleens were studded throughout with pinpoint yellow foci. Histological examination revealed that the yellow foci were bacterial colonies. Similar foci were also found in kidney, lung, heart, and muscularis of the intestine. Some of these colonies of bacteria were associated with a narrow peripheral zone of fibrin and necrotic cells, but there was no apparent cellular inflammatory reaction in most cases. *Y. pseudotuberculosis* type 1B was isolated from liver and spleen of both birds.

**11-73 and 12-73:** The frozen liver of one adult beaver and the frozen carcass of a second juvenile beaver were submitted on January 4. The first had been trapped, and the second found dead, both in the same area in Cochrane district. There were numerous 3-5mm white foci with irregular margins throughout all lobes of the liver in both cases, and also in the spleen of the second beaver. Histologically these foci were septic areas of caseous necrosis. An exudate was present on the capsule of liver and spleen of 12-73. Primary bacterial culture from the first case yielded a light mixed growth of *Staphylococcus*, *Streptococcus* and *Escherichia* spp.; there was no growth from the second case. Subsequent inoculation of guinea pigs in both cases resulted in abscessation of liver

within 9 days. Cultures of *Y. pseudotuberculosis* type 1B were isolated from these abscesses. The possibility of a latent infection in the guinea pigs was considered unlikely, since others from the same source, used for other experimental purposes, had showed no evidence of infection.

**430-73:** A juvenile beaver was found February 22 in the ice of South Crow Creek, Thunder Bay district. There were many small abscesses throughout the liver and intestine. Advanced decomposition precluded a histological examination. Bacterial cultures from the spleen and intestine yielded a heavy growth of *Y. pseudotuberculosis* type 1B.

Yersiniosis was considered the cause of abscesses in the trapped beaver and the apparent cause of death in all the other cases. The relationship between yersiniosis and pox in the crow was not determined.

#### ***Y. enterocolitica***

**431-73:** An adult beaver was found dead February 22 in the snow in the immediate vicinity of beaver 430-73. There were numerous abscesses throughout the liver and intestine. A histological examination was not undertaken. A heavy growth of *Y. enterocolitica* (biotype 1, phagetype 10o) was isolated from the intestine. The serological typing is not yet finalized. Yersiniosis was the apparent cause of death.

**2013-73:** An adult female raccoon was trapped on November 18 in Huron county. The raccoon was in good condition. Throughout the liver there were several well encapsulated nodules up to 3cm in diameter containing inspissated white exudate. Histologically these were septic abscesses surrounded by a thick zone of fibrous connective tissue. *Y. enterocolitica* (biotype 2, serotype 5,27, phagetype 10z) was isolated from a liver abscess and was the apparent cause of these abscesses.

**442-73:** One adult female Canada goose was found dead in Oxford county on April 11. The goose was in good flesh,

but the carcass was partially autolysed. There was a small amount of thick, orange fluid on the surface of Glisson's capsule on the right liver lobe. There was also a single 2mm metallic sphere, much harder than a lead shot, in the gizzard content. Subsequent toxicological analysis of hepatic tissue revealed only 0.3ppm lead. No other significant gross lesions were noted. Histological examination of liver revealed marked congestion, and a few lymphocytes and plasmacytes gathered around portal triads. The exudate noted at necropsy was a thick layer of autolysed erythrocytes on Glisson's capsule; the origin of this material was not determined. Lesions noted in the spleen were limited to congestion and a few irregular masses of hyaline material and lymphocytes suggestive of "reaction centers" of germinal follicles. A few colonies of both *Escherichia* and *Y. enterocolitica* (biotype 1, serotype 4,33, phagetype 10o) were isolated from splenic tissue, and a few colonies of *Escherichia* from hepatic tissue. There was no growth from the superficial hepatic exudate. Neither the cause of death nor the significance of the bacteria isolated were determined.

**881-73:** An adult male Pekin robin died suddenly at the Toronto Riverdale Zoo in September. The intestinal content was a frothy yellow fluid. There was white flocculent birefringent material on the serous membranes, the basis of a diagnosis of visceral gout. Post mortem change precluded a reliable microscopic examination, but there was no evidence of hepatic necrosis. A heavy growth of *Y. enterocolitica* (biotype 1, serotype 6,30, phagetype 10o) was isolated from the liver. The significance of the isolate was not determined. Another dead Pekin robin submitted at the same time did not yield *Yersinia*.

#### **Cases prior to 1973**

During the period from 1962 to 1972, *Y. pseudotuberculosis* had been isolated in this laboratory from four beavers and one snowshoe hare. There was no record of previous isolation of *Y. enterocolitica*.

## DISCUSSION

In most cases, our pathological findings are in agreement with those of previous authors who described abscesses in various tissues, primarily the liver and spleen, in both *Y. pseudotuberculosis* and *Y. enterocolitica* infections.<sup>19</sup> Also in agreement with our findings, Clark and Locke<sup>5</sup> reported enlargement of liver, spleen, and kidney in common grackles infected with *Y. pseudotuberculosis*, and yellowish areas on air sacs and serous surfaces. Langford<sup>12</sup> reported small necrotic foci in the mucosa of the intestine of various species infected with *Y. enterocolitica*. There are only two previous reports of *Y. enterocolitica* in birds. Langford<sup>12</sup> described post mortem lesions in the pigeons, and Halen and Meulemans<sup>8</sup> found lesions in the liver and spleen of the canary, but no characteristic lesions in the goldfinch. However, *Y. pseudotuberculosis* in addition to *Y. enterocolitica* was isolated from the liver of the canary. Since we found no abscesses in either the Canada goose or the Pekin robin, possibly birds may succumb to an acute enteritis and do not always exhibit a granulomatous reaction to *Y. enterocolitica* infection.

Mair<sup>11</sup> presented evidence for the transmission of *Y. pseudotuberculosis* from other animals to man, but stated that it has not been possible to establish a link between *Y. enterocolitica* infections in man and other animals. Similarly, Langford<sup>12</sup> states that the mode of transmission of *Y. enterocolitica* between either wild or domesticated animals, or these animals and man, is not known. However, evidence for the possibility of such transmission does exist. Dubois *et al.*<sup>6</sup> noted that cases of human yersiniosis in Belgium were most numerous in winter when wild hares, found to be carriers of *Y. enterocolitica*, are consumed. He speculated that since the same strains were not found in both species, they might be modified in passing from one to the other. Krogstad *et al.*<sup>10</sup> reported an outbreak of *Y. enterocolitica* type 2 in goats, and the occurrence of diarrhea in persons working with them. One of these persons had a high agglutinin titer against *Y. entero-*

*colitica* type 2, 1 week after experiencing diarrhea and abdominal pain. Ahvonen *et al.*<sup>1</sup> reported isolations of serotypes 3 and 9, common human strains in Europe, from two pigs, four dogs and one cat. Three of the isolates were from animal contacts of humans with verified *Y. enterocolitica* infection of the same serotype. Several other authors<sup>2, 7, 16, 17, 18, 21</sup> have reported serotypes 3 and 9 from pigs.

Toma<sup>18</sup> isolated *Y. enterocolitica* from oysters, and he suggests a possible etiological role of this organism in human gastroenteritis following meals of oysters. The serotypes isolated had been found in human infections. He also reported serotype 3 from both humans and a cat in Ontario, and five cultures of human origin and one from a dog that were serotype 5,27. This same serotype was isolated from a camel with diarrhea at the Metro Toronto Zoo (W. A. Rapley, personal communication), and from a raccoon in this laboratory.

In addition, the serotype isolated from the Pekin robin (serotype 6,30) was isolated by Toma from feces of four children with gastroenteritis in Ontario, and from two oysters from eastern Canada.

Lassen<sup>13</sup> expressed the opinion that the individual serotypes of *Y. enterocolitica* had their own characteristic reservoirs comprising only a very small number of species. Isolations of the same serotype from oysters, a Pekin robin, and humans; and another from a raccoon, a dog, a camel, and humans would appear to refute this, and to suggest a disease of zoonotic importance.

Hubbert<sup>9</sup> stated that there was as yet no evidence that birds were significant reservoirs of *Y. enterocolitica* or that they had a role in the spread of infection. Our recovery of this organism from a free-flying bird and a caged bird, in addition to other avian isolates,<sup>8,12</sup> indicates that as surveillance and recognition increases the presence of an avian reservoir may become more apparent. It is also likely that the non-avian wildlife reservoir for *Y. enterocolitica* is far greater than presently recognized.

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