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# DO NON-RUMINANT WILDLIFE POSE A RISK OF PARATUBERCULOSIS TO DOMESTIC LIVESTOCK AND VICE VERSA IN SCOTLAND?

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**ABSTRACT:** Paratuberculosis (Johne's disease) was long considered only a disease of ruminants. Recently non-ruminant wildlife species have been shown to harbor *Mycobacterium avium* subsp. *paratuberculosis*, the causative organism of paratuberculosis. We review the known non-ruminant wildlife host range of *M. avium* subsp. *paratuberculosis* and consider their role in the epidemiology of paratuberculosis in domestic ruminant livestock. *Mycobacterium avium* subsp. *paratuberculosis* has been isolated from lagomorph, canid, mustelid, corvid, and murid species. In agricultural environments domestic ruminants may contact wildlife and/or their excreta when grazing or feeding on farm-stored feed contaminated with wildlife feces, opening up the possibility of inter-species transmission. Of the wildlife species known to harbor *M. avium* subsp. *paratuberculosis* in Scotland, the rabbit is likely to pose the greatest risk to grazing livestock. Paratuberculosis in domestic ruminants is a notoriously difficult disease to control; the participation of non-ruminant wildlife in the epidemiology of the disease may partially account for this difficulty.

**Key words:** Johne's disease, *Mycobacterium avium* subsp. *paratuberculosis*, non-ruminant wildlife, paratuberculosis, transmission.

## INTRODUCTION

Paratuberculosis (Johne's disease) is a chronic enteritis caused by *Mycobacterium avium* subsp. *paratuberculosis*. Paratuberculosis was long considered a disease only of domestic and wild ruminants. The known host range of *M. avium* subsp. *paratuberculosis* recently has been extended to include non-ruminant wildlife species. The significance of paratuberculosis in non-ruminant wildlife populations has yet to be assessed and their potential role in the epidemiology of disease in domestic livestock is currently being investigated. Here we review the bacteriologic, pathologic, and epidemiologic features of paratuberculosis in non-ruminant wildlife and consider the potential for inter-species disease transmission in agricultural environments in Scotland (UK).

## PARATUBERCULOSIS IN WILDLIFE

Natural paratuberculosis in wild ruminants has been well documented, for ex-

ample in bighorn sheep (*Ovis canadensis*), tule elk (*Cervus elaphus nannodes*), white-tailed deer (*Odocoileus virginianus*), and bison (*Bison bison*) in the USA (Williams et al., 1979; Jessup et al., 1981; Chioldini and Van Kruiningen, 1983; Cook et al., 1997; Buergelt et al., 2000); roe deer (*Capreolus capreolus*) and red deer (*Cervus elaphus*) in Scotland (Sharp et al., 1995); and red deer and ibex (*Capra ibex*) in Italy (Ferroglia et al., 2000; Nebbia et al., 2000). Only recently has the natural occurrence of paratuberculosis in non-ruminant wildlife been described. A suspected isolation of *M. avium* subsp. *paratuberculosis* was first reported from a European brown hare (*Lepus europaeus*) in England (Matthews and Sargent, 1977) although the organism responsible was not fully characterized. Lesions attributed to paratuberculosis were subsequently described in a wild rabbit (*Oryctolagus cuniculus*) from Scotland (Angus, 1990). More re-

cently, presence of *M. avium* subsp. *paratuberculosis* was confirmed by the polymerase chain reaction (PCR) based species-specific IS900 insertion sequence detection method (Vary et al., 1990) in three surveys of rabbits from farms in the east of Scotland with a history of paratuberculosis in ruminant livestock (Greig et al., 1997, 1999; Beard et al., 2001b). Histopathologic changes varied from mild to severe (Beard et al., 2001b).

Following isolation of *M. avium* subsp. *paratuberculosis* in rabbits, studies were extended to other wildlife species on farms with a history of paratuberculosis in livestock. *Mycobacterium avium* subsp. *paratuberculosis* was isolated from foxes (*Vulpes vulpes*) and stoats (*Mustela erminea*) (Beard et al., 1999) and subsequently from weasels (*Mustela nivalis*), badgers (*Meles meles*), wood mice (*Apodemus sylvaticus*), rats (*Rattus norvegicus*), brown hares, jackdaws (*Corvus monedula*), rooks (*Corvus frugilegus*), and crows (*Corvus corone*) (Beard et al., 2001a).

Significance of *M. avium* subsp. *paratuberculosis* in non-ruminant wildlife is largely unknown. In wild ruminants, gross lesions and clinical signs are similar to those in infected cattle and sheep and the disease may be fatal (Williams et al., 1979; Buergelt et al., 2000). In contrast, mild lesions have been reported in non-ruminant wildlife. In rabbits, one study described thickened intestines and enlarged lymph nodes in three (10%) of 33 animals (Greig et al., 1997), while examination of a further 110 rabbits produced positive cultures from 17 and microscopic lesions in 18 rabbits (Beard et al., 2001b).

In foxes and stoats, *M. avium* subsp. *paratuberculosis* was isolated from samples of intestines and mesenteric lymph nodes. Histopathologic changes similar to those described in ruminants with subclinical paratuberculosis were noted in these tissues. These changes were more subtle than those seen in either advanced ruminant paratuberculosis or severely infected rabbits (Beard et al., 2001a, b). In crows,

rooks, jackdaws, rats, wood mice, and hares, *M. avium* subsp. *paratuberculosis* was isolated from intestinal and lymphoid tissue with few or no microscopic lesions (Beard et al., 2001b). The findings of Beard et al. (2001a) make it clear that non-ruminant species can harbor *M. avium* subsp. *paratuberculosis* and that the natural host range is far greater than previously thought.

Transmission of paratuberculosis in livestock is believed to occur mainly through the fecal–oral route (Clarke, 1997), i.e., through ingestion of infected feces or fecal contaminated feed. Below we detail types of contact between wildlife and domestic ruminant livestock that may represent routes of disease transmission.

#### POTENTIAL TRANSMISSION ROUTES TO WILDLIFE

Isolation of *M. avium* subsp. *paratuberculosis* from non-ruminant wild species on farms with paratuberculosis in livestock, suggests that interspecies transmission may occur between livestock and non-ruminant wildlife. Interspecies transmission has been demonstrated experimentally; rabbits were infected with a bovine strain of *M. avium* subsp. *paratuberculosis* (Mokresh and Butler, 1990). Sheep and feral goats became infected after sharing grazing pasture with cattle infected with paratuberculosis (Ris et al., 1987, 1988). Given that clinically infected cattle and sheep shed in excess of  $10^8$  bacilli per gram of feces (Cranwell, 1997; Whittington et al., 2000), it is likely that wild herbivores such as rabbits or hares sharing pasture with infected cattle could pick up bacteria through the ingestion of fecal contaminated vegetation.

Based on a sample of 20 farms across Scotland, Greig et al. (1999) found a statistically significant relationship between farms with previous or current paratuberculosis in cattle and the presence of paratuberculosis in rabbits. Furthermore, on the basis of genetic typing, no differences were found between the strains of *M. av-*

*ium* subsp. *paratuberculosis* isolated from rabbits and cattle, suggesting that transmission between the species had occurred (Greig et al., 1999).

Isolation of *M. avium* subsp. *paratuberculosis* in granivorous or omnivorous rodents is more likely to be explained by their presence in buildings housing infected livestock than from fecal contaminated pasture (Beard et al., 2001a). Indeed all rodents from which *M. avium* subsp. *paratuberculosis* was isolated were caught in buildings which housed cattle, whereas those captured in adjacent field margins or woodland were negative on culture (Daniels and Hutchings, unpubl. data). Rodents may become infected through scavenging livestock feed on floors contaminated with livestock feces.

Carnivores, such as stoats, foxes, and weasels may become infected with *M. avium* subsp. *paratuberculosis* through ingestion of infected prey. *Mycobacterium avium* subsp. *paratuberculosis* was isolated from mesenteric lymph nodes and intestinal tissue of infected rabbits (Greig et al., 1997, 1999). Foxes, stoats, and weasels ingest entire rodents and a sufficient proportion of rabbit carcasses to include potentially infected lymph or intestine tissue. Lagomorphs and small rodents form the majority of the diet of foxes, stoats, and weasels in Scotland (Harris and Lloyd, 1991; King, 1991a, 1991b; Leckie et al., 1998).

That predation is a mode of transmission to carnivores is further suggested by the higher prevalence in predator compared to prey species. The overall prevalence in predators was 62% (23/27 foxes, 17/37 stoats, and 2/4 weasels) compared to 10% for prey species (14/83 rabbits, 3/35 rats, and 3/88 wood mice). Transmission of pathogens through predation has been reported and is explained by carnivores' higher trophic level in the food chain (e.g., Zarnke et al., 2000).

At least part of the diet of predators and a large part of the diet of corvids may consist of scavenging potentially infected prey

carcasses (Mason and Macdonald, 1995). Therefore, scavenging may constitute an additional mode of paratuberculosis transmission. Corvids also feed in buildings housing livestock and prey on invertebrates associated with livestock pasture.

#### POTENTIAL TRANSMISSION ROUTES FROM WILDLIFE TO LIVESTOCK

The high prevalence of paratuberculosis in some non-ruminant wildlife species and their interaction with susceptible ruminant livestock raises the possibility that they play a role in the epidemiology of the disease in domestic livestock. The risks of transmission from wildlife to livestock have been raised frequently, but they are difficult to prove in the field mainly due to the long incubation period of the disease and the difficulty in excluding other potential sources of infection (Williams et al., 1979; Chiodini and Van Kruiningen, 1983; Greig et al., 1997; Beard et al., 1999, 2001a, b; Ferroglio et al., 2000).

Experimentally, calves were infected with a *M. avium* subsp. *paratuberculosis* strain from a wild rabbit (Beard et al., 2001c). Also, a study of four farms in Scotland with a history of paratuberculosis in wild rabbits and domestic livestock found that infected rabbits excreted up to  $4 \times 10^6$  colony forming units per gram of feces, implying that the ingestion of sward contaminated with just one or two infected rabbit fecal pellets would theoretically be sufficient to constitute an infective dose (Hutchings et al., in press a). Rabbits utilize improved agricultural pastures for grazing and for burrows at high density and thus these pastures become contaminated with their feces (Daniels et al., in press). Rates of contamination of agricultural pastures on the same four farms was 7,357 rabbit fecal pellets/ha/day (Hutchings et al., in press a). Domestic ruminants grazing fecal contaminated pasture is a common route of intra- and inter-species disease transmission (Hutchings et al., in press b). Cattle do not avoid grazing rabbit fecal contaminated pasture (Daniels et al.,

2001), unlike the avoidance behavior they show towards their own feces (Bao et al., 1998) and carnivore feces (Hutchings and Harris, 1997). Therefore, in Scotland the combination of high prevalence and level of infection in rabbits and their feces, high levels of fecal contamination of agricultural pastures by rabbits, and the lack of avoidance by cattle to grazing pastures contaminated with rabbit feces, suggests that rabbits represent a risk of *M. avium* subsp. *paratuberculosis* transmission to grazing domestic livestock. This risk from rabbits is likely to be greatest for the more susceptible young livestock (Sweeney, 1996).

Livestock may also be at risk of infection from feed contaminated with wildlife feces (Daniels and Hutchings, 2001). Beard et al. (2001a) obtained positive cultures from feces of rats and wood mice captured in buildings housing livestock and/or livestock feed. Significant quantities of rodent feces may contaminate livestock feed stored on the ground in farm buildings (with a mean of 80 fecal pellets per m<sup>2</sup> of stored feed deposited per mo). Livestock presented with feed contaminated with rodent feces ingest a high proportion (up to 100%) of the feces encountered (Daniels and Hutchings, 2001), suggesting that this also represents a potential mode of transmission. However, to date disease prevalence recorded in rodent species is low relative to rabbits and their predators.

The importance of carnivores as wildlife hosts of paratuberculosis is more likely to be indirect, because livestock are unlikely to come into contact with large amounts of their feces. Corvids, on the other hand, may occur on farms at relatively high densities and so cannot be ruled out as a source of infection for domestic livestock. Perhaps more importantly infected carnivores and corvids could theoretically transport *M. avium* subsp. *paratuberculosis* over far greater distances than prey species. For example, in the east of Scotland the home range size of rabbits is 6.3 ha (Hulbert et al., 1996) but 4,000 ha for fox-

es (Harris and Lloyd, 1991). Predators and scavengers of rabbits may thus play a role in the spatial spread of the disease.

Paratuberculosis in domestic livestock populations is a notoriously difficult disease to control (Stabel, 1998). Non-ruminant wildlife hosts of *M. avium* subsp. *paratuberculosis* may partially account for these difficulties. We are at an early stage in understanding the role of wildlife in the ecology of paratuberculosis. For example, the relative force of infection of *M. avium* subsp. *paratuberculosis* from wildlife to domestic livestock and vice versa is currently unknown. This review has highlighted the potential for non-ruminant wildlife to play a role in the ecology of paratuberculosis. However, further research effort is needed if we are to quantify that role. Specifically, in relation to wildlife, further studies are needed to determine the dynamics of disease in wildlife populations and particularly whether any non-ruminant wildlife species represent true reservoirs of disease.

## CONCLUSIONS

The recent findings of a wide host range of *M. avium* subsp. *paratuberculosis* among non-ruminant wildlife species on farms with paratuberculosis in livestock has important implications for an understanding of the disease, both in wildlife and in livestock. Potential transmission routes to wildlife include predation, scavenging, and sympatry with infected livestock. Conversely, transmission to livestock from non-ruminant wildlife could be a factor in the epidemiology of paratuberculosis, with important implications with respect to attempted control or eradication of the disease. All current evidence from Scotland suggests that, of the wildlife species known to harbor *M. avium* subsp. *paratuberculosis*, the rabbit poses the greatest risk of transmission to domestic livestock.

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