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Estimation of flock/herd-level true *Mycobacterium avium* subspecies *paratuberculosis* prevalence on sheep, beef cattle and deer farms in New Zealand using a novel Bayesian model

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ABSTRACT

The study aimed to estimate the national- and island-level flock/herd true prevalence (HTP) of *Mycobacterium avium* subsp. *paratuberculosis* (MAP) infection in pastoral farmed sheep, beef cattle and deer in New Zealand. A random sample of 238 single- or multi-species farms was selected from a postal surveyed population of 1940 farms. The sample included 162 sheep flocks, 116 beef cattle and 99 deer herds from seven of 16 geographical regions. Twenty animals from each species present on farm were randomly selected for blood and faecal sampling. Pooled faecal culture testing was conducted using a single pool (sheep flocks) or two pools (beef cattle/deer herds) of 20 and 10 samples per pool, respectively. To increase flock/herd-level sensitivity, sera from all 20 animals from culture negative flocks/herds were individually tested by Pourquier® ELISA (sheep and cattle) or Paralisa™ (deer). Results were adjusted for sensitivity and specificity of diagnostic tests using a novel Bayesian latent class model. Outcomes were adjusted by their sampling fractions to obtain HTP estimates at national level. For each species, the posterior probability (POPR) of HTP differences between New Zealand North (NI) and South (SI) Islands was obtained.

Across all species, 69% of farms had at least one species test positive. Sheep flocks had the highest HTP estimate (76%, posterior probability interval (PPI) 70–81%), followed by deer (46%, PPI 38–55%) and beef herds (42%, PPI 35–50%). Differences were observed between the two main islands of New Zealand, with higher HTP in sheep and beef cattle flocks/herds in the NI. Sheep flock HTP was 80% in the NI compared with 70% (POPR = 0.96) in the SI, while the HTP for beef cattle was 44% in the NI and 38% in the SI (POPR = 0.80). Conversely, deer HTP was higher in the SI (54%) than the NI (33%, POPR = 0.99). Infection with MAP is endemic at high prevalence in sheep, beef cattle and deer flocks/herds across New Zealand.

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1. Introduction

Paratuberculosis (Ptb), caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP), is a chronic granulomatous enteric disease, which occurs worldwide and affects domestic ruminant species including deer, sheep, and cattle (Harris and Barletta, 2001). A large proportion of infected animals will remain subclinically infected during their entire productive life, with no measurable effect on production (Nielsen and Toft, 2008). The isolation of MAP from intestinal tissue of Crohn's disease (CD) patients, a human chronic inflammatory bowel disease, has suggested a possible link between the two diseases, with MAP-contaminated animal produce a possible source of CD in humans (Mishina et al., 1996; Feller et al., 2007). The apparent association between MAP in animals and humans has raised concerns within the agricultural industry about the public health relevance of MAP, and the potential effect of high MAP prevalence on trade or consumption of milk and meat.

The first Ptb case in New Zealand was recorded in 1912 in an imported cow (de Lisle, 2002). The disease was first reported in New Zealand sheep in 1952, and in the 1980s Ptb was confirmed in farmed deer (de Lisle et al., 1993). Despite MAP infection being thought to be endemic in the New Zealand, no population-based estimates of MAP infection prevalence or disease incidence are currently available for sheep, beef or dairy cattle. Conversely, in deer, a national herd-level true prevalence (HTP) of 59% has been estimated, based on tissue culture of randomly selected lymph nodes from abattoir surveillance (Stringer et al., 2013b). Moreover, a significant prevalence difference was reported between New Zealand North (NI) and South (SI) Islands, 44% and 67% respectively. However, no farm-based estimate is available for this species. Infection prevalence estimation is a key element to assess the disease impact, and for the design of control programmes. MAP prevalence has been previously estimated in other countries using Bayesian latent-class modelling (Sergeant and Baldock, 2002; Nielsen et al., 2007; Dhand et al., 2010; Okura et al., 2010). This methodology has the advantage that adjustments can be made for sensitivity and specificity of diagnostic tests, in a flexible probabilistic framework, allowing researchers to model complex testing designs to obtain true prevalence estimates.

In New Zealand, domestic ruminants are commonly farmed in multi-species pastoral systems, where sheep, beef cattle and/or deer are often grazed on the same pastures, either concurrently or successively. Therefore, Ptb research in New Zealand addresses all species in an overarching strategy, in order to develop an integrated approach to Ptb control (JDRC, 2011). The objective of this study was to estimate HTP of MAP infection in sheep, beef cattle and deer flocks/herds in New Zealand, at national and island level (North Island (NI) and South Island (SI)). The study also aimed to assess the distribution of the infection in the populations, and to explore/validate prevalence differences between islands, as it has previously been reported for deer herds.

2. Materials and methods

2.1. Selection of farms

Farms were selected in two stages. In the first stage (*a*), from December 2008 to March 2009 a postal survey was mailed out to 7998 farmers. The survey simultaneously gathered information about two different diseases (Ptb and leptospirosis). The contacted farmers were clients of 28 large animal veterinary practices, located in four administrative regions in the NI (Waikato, Wairarapa, Hawkes Bay, Manawatu-Wanganui), and three in the SI (Marlborough, Canterbury, Southland). The survey targeted 'commercial' sheep, deer, and/or beef cattle operations (i.e. those with a minimum of 40 deer, 400 sheep, and/or 40 beef cattle). In New Zealand, there are 24,644 farms meeting these criteria with at least one of the three species under study, based on 2009 Agribase™ data (national population database). The questionnaire gathered retrospective information about animal demographics, reproduction performance, Ptb & leptospirosis incidence, and grazing management information on all ruminant species present on the farm. Those data are reported elsewhere (Verdugo et al., 2010). Correctly completed questionnaires were returned from 1940 (24.3%) commercial single- or mixed-species farms, constituting the sampling frame (source population). In the second stage (*b*), farms were randomly selected from the source population with the aim of sampling of one group of each ruminant species present (sheep, beef cattle and/or deer).

2.2. Sampling protocol and laboratory testing

Sampling was carried out by contracted veterinary practitioners from June 2009 to July 2010. Twenty animals from each species present on farm were randomly selected.[1] Paired faeces and serum samples were collected from sheep (ewes, 2-years and older), beef cattle (cows, 2-years and older) and deer (yearlings, i.e. 12–24 months, either sex). A single pool was prepared from sheep faeces (20 samples/pool), and two pools were prepared from beef cattle and deer (10 samples/pool) from each farm. Culture was performed by the Wallaceville Animal Health Laboratory, Upper Hutt, using BACTEC 12B liquid culture medium containing egg yolk and mycobactin, after a decontamination step with cetylpyridinium chloride, as described by Whittington et al. (1999). If a pooled faecal culture (PFC) was positive, the entire flock/herd was classified as being positive. Individual serum samples from culture negative flocks/herds only were tested by ELISA tests: Pourquier® ELISA for sheep and cattle (Institut Pourquier, Montpellier, France), and Paralisa™ for deer (Griffin et al., 2005). In a given species, a flock/herd was defined as positive if a PFC pool or ELISA test (group level interpretation, cut off = 1 + ve animal) was positive.

[1] Animals belonging to the target age group were rounded up and yarded. Then, animals were selected using a systematic random sampling procedure as they were released from the yard.