



## Spatial differences in occurrence of paratuberculosis in Danish dairy herds and in control programme participation

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### ABSTRACT

Paratuberculosis is a chronic infection of economic importance to the cattle industry and a voluntary control programme is offered to Danish dairy farmers. Our objective was to evaluate spatial differences in both control programme participation and paratuberculosis prevalence in Denmark.

The study included 4414 dairy herds: 1249 were participating in the control programme, and 1503 were tested for antibodies to *Mycobacterium avium* subsp. *paratuberculosis* (MAP). Spatial differences were evaluated by kernel smoothing, kriging, and cluster analysis. Participation was lowest among herds on the island Zealand ( $\leq 23\%$ ). The risk of a herd being infected with MAP was found to be high on most of Zealand, but the uncertainty of this result was large due to a limited number of tested herds. In the rest of the country, the south western part of the peninsula Jutland had the highest risk of MAP ( $\geq 91\%$ ). The risk of MAP was also high (86–91%) in the northern part of both Jutland and Funen. The predicted apparent within-herd prevalence was highest (5–8.5%) in some local areas across Jutland, in the north western part of the island Funen, and in the south and western part of Zealand. Scan statistics located the primary cluster of herds with high apparent within-herd prevalence in the western part of Funen. Furthermore, a number of significant clusters were found in Jutland and a single significant cluster in Zealand. Consistency was found between kriging and scan statistics results with respect to location of areas with high apparent within-herd prevalence of MAP.

Potential explanations for differences in participation include herd size and local herd health advisers, whereas for example soil characteristics might influence prevalence. Further studies are needed to evaluate these and other risk factors.

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### 1. Introduction

Paratuberculosis is a chronic infection in cattle and other ruminants caused by *Mycobacterium avium* subsp. *paratuberculosis* (MAP). The main clinical symptoms of MAP infection are diarrhoea and weight loss (Chiodini et al., 1984). Furthermore, infection causes reduced milk yield and eventual death of infected animals (Nielsen, 2009,

chap. 1), all leading to economic losses to the farmers. MAP is also suspected of being involved in the development of Crohn's disease in humans (Behr and Kapur, 2008). Infectious animals shed MAP in faeces and thereby contaminate the environment resulting in transmission of MAP (Sweeney, 1996). MAP can also be transmitted in milk and colostrum from infectious animals (Streeter et al., 1995), and *in utero* from dam to calf (Whittington and Windsor, 2009).

It has been estimated that approximately 80–85% of all Danish herds are infected with MAP (Nielsen, 2009). A voluntary control programme for MAP in Danish dairy

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herds was established by the Danish Cattle Federation in 2006 (Nielsen, 2007). The programme aims at reducing the prevalence of MAP and providing farmers with tools to manage MAP infection. All lactating cows in herds participating in the programme are automatically tested for MAP four times a year using a milk antibody ELISA. Since only lactating cows are tested, most cows are tested approximately three times a year. At each screening, approximately 89% (std. dev. = 5.5%) of all cows within a herd are tested. Dry cows are not tested. By January 2009, approximately 28% of Danish dairy herds participated in the programme (Nielsen, 2009, chap.7).

The spatial distribution of participation in the voluntary control programme and of MAP infection in Denmark has never been examined. Hence, the objectives of this study were to (a) describe the spatial pattern of participation in the voluntary control programme for MAP and (b) describe the spatial pattern of apparent MAP prevalence throughout Denmark, including identification of areas with increased risk of MAP.

## 2. Materials and methods

### 2.1. Dairy herds included in the study

All Danish cattle herds are registered in The Danish Central Husbandry Register with a unique identification number. From this register, information on dairy herds were extracted. To be eligible for inclusion in this study a dairy herd had to have been producing milk during the first quarter of 2009 ( $n_{herd} = 4462$ ). Information included herd location given as UTM EUREF89, zone 32 coordinates. Coordinates are based on location of the largest building on the premises. Herds with missing information for location ( $n_{herd} = 1$ ) were excluded, as were herds located on the remote Danish island of Bornholm ( $n_{herd} = 47$ ). For this analysis, dairy herds were considered participants in the voluntary control programme if they were listed as participating on January 1, 2009. Enrolled herds perform 4 annual herd screenings. Samples are automatically collected through the Danish milk recording system on 4 test dates at regular intervals. Enrolled farmers therefore can not avoid being tested on these test dates, unless the milk samples are lost at the laboratory. Non-enrolled herds may also undertake partial or complete herd screenings on their own initiative. A total of 4414 dairy herds were included in the study of participation in the control programme, with 1249 herds participating in the programme.

All tests for MAP were performed and recorded at the individual cow level. By October 15, 2008, a new test (ID-Screen<sup>®</sup>, ID-Vet, Montpellier, France) was introduced. Only data from this new test were used. All available data during the study period from October 2008 to June 2009 were used, even though some herds ( $n_{herd} = 311$ ) were not officially enrolled in the control programme at the time of testing. Only one date of testing within each herd was used. For each herd, this was done by randomly choosing one test date during the last three months of 2008 or, if no testing took place during that period, by including the first test in 2009. Herds with a number of tested cows less than 10 ( $n_{herd} = 47$ ) were excluded. By visual inspection, excluded

herds seemed to be randomly located across Denmark. Some herds had been vaccinated against MAP and were therefore also excluded ( $n_{herd} = 8$ ). A total of 1503 herds were tested for MAP. A total of 254 herds had therefore been tested without being participating in the programme on January 1, 2009.

A cow was defined to be test-positive if the ELISA test had a sample-to-positive ratio of 0.30, corresponding to manufacturer's recommendations (ID Vet, Montpellier, France). The specificity has been estimated to 0.98 in the programme (Nielsen, unpublished data) and the sensitivity varies with age of infection (0.06 at 2 years of age to 0.50 at 5 years of age) (Nielsen and Toft, 2006). If at least one cow within a herd was test-positive, the herd was defined to be MAP-infected. The apparent within-herd prevalence of MAP was calculated as the number of test-positive cows in the herd divided by the number of tested cows in the herd.

### 2.2. Spatial analyses

Consider locations of dairy herds participating in the control programme a spatial point process with intensity function  $\lambda_1$  and locations of non-participating dairy herds a spatial point process with intensity function  $\lambda_0$ . Then the probability of a herd at a given location  $s$  participating in the control programme can be estimated as

$$p(s) = \frac{\lambda_1(s)}{\lambda_0(s) + \lambda_1(s)}$$

The spatial pattern of participation in the control programme was described by mapping a smoothed estimate of  $p(s)$ . The estimate was calculated on a grid of 5 km  $\times$  5 km grid cells by estimating the intensity functions and calculating the ratio. The intensity function  $\lambda$  of a spatial point process  $z = (z_1, z_2)$  was estimated by a non-parametric kernel estimator (Diggle, 1985)

$$\hat{\lambda}_h(s) = e_h(s) \sum_{i=1}^n \frac{1}{h^2} \kappa\left(\frac{s - z_i}{h}\right)$$

where  $h$  is the bandwidth,  $\kappa$  is a kernel function chosen to be Gaussian, and  $e_h(s)$  is an edge correction factor given by

$$e_h(s) = \int_A \frac{1}{h^2} \kappa\left(\frac{s - u}{h}\right) du$$

where  $A$  is the sample region. The performance of the estimator is mainly determined by the bandwidth  $h$  and not the choice of kernel function. The Gaussian optimal method (Bowman and Azalini, 1997) was used to estimate the bandwidth for each of the two intensity functions  $\lambda_0$  and  $\lambda_1$ . The bandwidth was structured as  $h = (\tilde{h} \times \text{std.dev.}(z_1), \tilde{h} \times \text{std.dev.}(z_2))$ . Based on the estimated bandwidth, a common value was chosen for estimating  $\lambda_0$  and  $\lambda_1$ . To take into account the much lower herd density on Zealand (Fig. 1a), Denmark was divided into two regions (Zealand and the rest of the country) when estimating bandwidth and hence intensity functions.

The spatial pattern of the risk of a herd being infected was described in a similar manner by letting  $\lambda_0$  be the

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