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Title

Bayesian estimation of prevalence of paratuberculosis in dairy herds enrolled in a voluntary Johne's Disease Control Programme in Ireland.

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Abstract

Bovine paratuberculosis is a disease characterised by chronic granulomatous enteritis which manifests clinically as a protein-losing enteropathy causing diarrhoea, hypoproteinaemia, emaciation and, eventually death. Some evidence exists to suggest a possible zoonotic link and a national voluntary Johne's Disease Control Programme was initiated by Animal Health Ireland

in 2013. The objective of this study was to estimate herd-level true prevalence (HTP) of paratuberculosis in Irish herds enrolled in the national voluntary JD control programme during 2013-14. Two datasets were used in this study. The first dataset had been collected in Ireland during 2005 (5,822 animals from 119 herds), and was used to construct model priors. Model priors were updated with a primary (2013-14) dataset which included test records from 99,101 animals in 1,039 dairy herds and was generated as part of the national voluntary JD control programme. The posterior estimate of HTP from the final Bayesian model was 0.23 - 0.34 with a 95% probability. Across all herds, the median animal-level true prevalence was found to be 0.032 (0.009, 0.145). This study represents the first use of Bayesian methodology to estimate the prevalence of paratuberculosis in Irish dairy herds. The HTP estimate was higher than previous Irish estimates but still lower than estimates from other major dairy producing countries.

"Paratuberculosis"; "Dairy"; "Ireland"; "Prevalence"; "Bayesian"

1. Introduction

Bovine paratuberculosis is a disease characterised by chronic granulomatous enteritis which manifests clinically as a protein-losing enteropathy causing diarrhoea, hypoproteinaemia, emaciation and, eventually death (Sweeney, 2011). Adverse effects on animal productivity in terms of lower milk yield, higher cull rates, reduced value for culled animals, possible adverse effects on fertility and losses due to continued spread of infection are key drivers in the attempt to control the disease at farm level. In addition some research exists to suggest that the aetiologic pathogen *Mycobacterium avium* subspecies *paratuberculosis* (MAP) may pose a zoonotic risk (Chiodini et al., 2012). Consequently, many major dairy producing countries have introduced control programmes aimed at reducing overall prevalence (Geraghty et al., 2014).

Animal Health Ireland (AHI) was formed as a not-for-profit organisation providing national leadership and coordination of non-regulatory animal health issues in Ireland (More et al., 2011). The AHI Johne's Disease Control Programme was developed and introduced as a

49 voluntary programme in 2013. Irish herd-level true prevalence (HTP) on dairy farms in 2005
50 was estimated at 20%, based on the results of a serological survey (Good et al., 2009),
51 considerably lower than estimates across Europe of greater than 50% (Nielsen and Toft, 2009).
52 In common with trends across the EU, the number of dairy herds in Ireland has been gradually
53 decreasing whilst herd sizes have increased. It is therefore possible that HTP has altered in the
54 intervening years.

55 Measuring the impact of control programmes requires an initial baseline estimation of the
56 occurrence of infection. In the context of chronic diseases of slow or insidious onset such as
57 paratuberculosis, incidence may be difficult to calculate and prevalence is often used instead
58 (Messam et al., 2008). A review of the prevalence of paratuberculosis across countries in Europe
59 identified critical issues in a number of studies (Nielsen and Toft, 2009), primarily these issues
60 related to the incorrect values for test sensitivity (Se) and specificity (Sp) in the analysis.

61 Estimates of Se and Sp of diagnostic tests for paratuberculosis vary considerably (Nielsen and
62 Toft, 2008). Much of this variation can be attributed to differences among reference populations
63 and sampling strategies that have been used for the test validation procedure (Greiner and
64 Gardner, 2000). However estimates of Se and Sp may also vary according to prevalence
65 (Brenner and Gefeller, 1997) and therefore between herds (Greiner and Gardner, 2000).
66 Consequently, the relationship between true prevalence (TP) and apparent prevalence (AP) can
67 be expected to vary between populations. It may therefore be unreasonable to assume a fixed,
68 constant, Se and Sp over different populations (Berkvens et al., 2006). In Bayesian analyses, all
69 parameters are considered random variables and can be modelled using probability
70 distributions. Uncertainty and variability associated with estimates of test Se and Sp may
71 therefore be incorporated in the analysis. In addition, in this instance, a Bayesian posterior
72 probability will provide inference on a prevalence estimate, conditional on both currently
73 observed data and previous information about the disease. This methodology has not yet been
74 applied to the estimation of the prevalence of paratuberculosis in Irish dairy herds, but has been

used extensively to estimate the prevalence in other countries (Pozzato et al., 2011; Lombard et al., 2013; Verdugo et al., 2015)

The aim of this study, therefore was to estimate the HTP and overall animal-level true prevalence (ATP) of paratuberculosis among herds enrolled in a national voluntary control programme.

2. Materials and Methods

Two datasets were analysed in this study. The primary analysis utilised test data collected from the national control programme between 2013 and 2014. Model priors for this analysis were constructed by analysing a secondary (2005) dataset.

2.1. Study Population

The primary (2013-2014) dataset for the current study was obtained from herds voluntarily enrolled in the national voluntary Johne's Disease control programme. Herds enrolled in the voluntary programme are required to have all animals that are 24 months of age and older serologically tested using either serum or milk samples. Diagnostic testing is conducted in both government and commercial laboratories using one of 3 commercial ELISA kits approved for use in the AHI programme; Parachek, Prionics, Switzerland (kit A), Paratuberculosis Antibody Screening Test, Idexx, USA (kit B) and ID Screen, IDVet, Montpellier, France (kit C). Producers that elect to test using blood or milk sample are required to test all eligible animals once or twice per year respectively. Test data, including follow up testing, are stored centrally in the Irish Cattle Breeding Federation computer database. Data were extracted for the period beginning 1st November 2013 and ending 30th December 2014 and included anonymised cow and herd identifiers, test-date, sample-to-positive (S/P) ratio, laboratory interpretation (negative, suspect, positive), sample type (blood or milk), testing laboratory (test kit) and county.

99 Test data also included follow up testing data on subsamples of animals within herds. Herd test
100 data were available for 1,040 herds, 436 of these had conducted 2 or more additional rounds of
101 testing. In order to avoid bias that may have been introduced by some herds conducting greater
102 than 1 herd screen, only one test per animal was used. The first recorded test result for each
103 animal was used for the purpose of this analysis and Se and Sp values were based on a single
104 test strategy. The "herd" in this study was therefore defined as the number of unique and
105 eligible animals on the farm within the 14 month sampling frame.

106 **2.2. Statistical Analysis**

107 **2.2.1. Analytical model**

108 Prevalence was estimated with a Bayesian model extended from that proposed by Branscum
109 (2004), which was based on methodology introduced by Hanson (2003). The number of animals
110 testing positive in each herd was considered to be binomially distributed. A binomial rather
111 than a hypergeometric distribution was used because all adult animals in each herd were
112 sampled. The model was constructed as;

113 $npos_i \sim \text{Binomial}(\pi_i, nherd_i)$ (1)

114 $\pi_i = Se_{jk} \times ATP_i + (1\text{-}ATP_i) \times (1\text{-}Sp_{jk})$ (2)

115 $ATP_i = HTP_i \times CWHP_i$ (3)

116 $HTP_i \sim \text{Bernoulli}(\mu)$ (4)

117 $CWHP_i \sim \text{Beta}(a_{CWHP}, b_{CWHP})$ (5)

118 $Se_{jk} \sim \text{Beta}(a_{Se}, b_{Se})$ (6)

119 $Sp_{jk} \sim \text{Beta}(a_{Sp}, b_{Sp})$ (7)

120 $\mu \sim \text{Beta}(a_\mu, b_\mu)$ (8)

where $npos_i$ equals the number of animals testing positive in the i-th herd ($herd_i$), given a probability of each animal testing positive (π_i) and number of animals in the herd ($nherd_i$). The probability of a randomly chosen animal from a herd testing positive was a function of the animal-level true prevalence (ATP) within $herd_i$, and the diagnostic test characteristics; Se and Sp, which varied according to kit (j) and test medium (k). The ATP for a given herd was modelled as a mixture distribution: the product of HTP and conditional-herd prevalence (CWHP). The HTP was modelled as a Bernoulli distribution. The Bernoulli distribution is used to model random variables with two possible outcomes, in this case a herd was considered to be "infected" with probability μ to indicate the probability of a randomly chosen herd containing one or more truly infected animals and "uninfected" with a probability 1-μ. Then, conditional on the herd being infected, the conditional within-herd prevalence (CWHP) was modelled as beta distribution. Beta distributions are a relatively flexible family of distributions on the real number line from 0-1 and are a common method of modelling prevalence.

The effect of ELISA kit and test medium used was assessed using random and fixed effects, however the change in the animal-level apparent prevalence due to the effect of these variables was found to be low (<0.005) and they were removed again from the model.

2.2.2. Model Priors – Test Characteristics

Nielsen and Toft (2008) proposed the case definitions "infected", "infectious" and "affected" in an attempt to reduce variability between reported estimates of test Se. The subgroup "infected" also includes animals that are "infectious" and "affected", and is the population of interest in this prevalence study.

To estimate the Se and Sp of each commercial kit, a published review of the literature (Nielsen and Toft, 2008) was examined and supplemented with searches in PubMed and CABdirect of all literature published between 2007 and 2015 on paratuberculosis diagnostic test evaluation. Test characteristics for each test kit used in Ireland evaluating the "infected" sub group, were

146 extracted from each peer-reviewed article from this search and from the 2008 review
147 publication (Table 1).

148 The first study was limited to a population of cull cows (McKenna et al., 2005) and the second
149 study (Norton et al., 2010) was carried out on herds with a history of clinical disease and with
150 relatively high ATP. A third study (Nielsen et al., 2013), was removed because the target
151 condition "infected", was in this case, defined based on the longitudinal interpretation of the
152 evaluated serological test. A final study (Aly et al., 2014) was removed which was based on the
153 evaluation of the test on a single herd.

154 After removing these estimates, 2 evaluation studies were available for kit A with no
155 appropriate published values available for kits B and C. When test characteristics were
156 presented by age group, a weighted mean of the test Se was calculated relative to the age
157 distribution of the present study. A sample size weighted mean was next calculated for the Se of
158 kit A (0.224) using the two estimates extracted from the study. A previously constructed
159 estimate for the Se and Sp of kit B was available (Nielsen and Toft, 2009) which has been used in
160 subsequent prevalence estimates (Pozzato et al., 2011), kits B and C are known to have similar
161 ancestry, therefore the same values were adopted for kit C. The parameters for the beta-
162 distribution were found using "betabuster" software (Chun-Lung 2010) based on a given mode
163 and either upper or lower 95th bound. The Se of individual milk ELISA relative to serum ELISA
164 has been shown to be approximately 0.87 (van Weering et al., 2007), therefore in the absence of
165 a Se estimate for milk, the Se of the serum ELISA was multiplied by a factor of 0.87. Final values
166 and associated beta distribution parameters are shown in Table 2.

167 **2.2.3. Model priors – HTP and CWHP**

168 Prior distributions for HTP and CWHP in Irish dairy herds were required. In order to construct
169 these priors, data (secondary dataset) from a previously published prevalence survey (Good et
170 al., 2009) were used as follows. Data were removed from animals less than 24 months of age,
171 from animals without a recorded date of birth and from non-dairy enterprises. This dataset

included a much higher proportion of small herds relative to the primary dataset, therefore, farms containing less than 20 animals were removed to prevent possible overestimation of CWHP priors due to small herd sizes.

The CWHP was estimated for each positive herd using the Rogan-Gladen estimator (Rogan and Gladen, 1978), i.e., CWHP = (AP+Sp-1)/(Se+Sp-1), where, AP = Apparent Prevalence. All serum samples in this survey were tested using the Pourquier ELISA, this kit is now sold as Kit B, and therefore, the test characteristics given for Kit B (Table 2) were used to calculate the prior distribution of within-herd prevalences. The distribution of CWHPs in this dataset were plotted and the mean and mode used to fit a beta distribution using the betabuster programme.

A number of priors were trialled for HTP including the herd-level apparent prevalence based on a varying number cut point reactors. However, after it was observed that the primary model was extremely insensitive to the prior for HTP, it was decided to use a flat distribution from 0 - 1 as the prior for this variable.

2.2.4. Sensitivity Analysis

Sensitivity analysis of the final estimate to the priors used in the model was assessed by varying the point estimate and confidence intervals of the each prior by 10%, 25% and 50% in either direction and repeating the analysis. In addition, the prior for HTP was modelled as a uniform distribution from 0 – 1 and the analysis repeated. The posterior HTP was compared with the estimate from the default priors and the percentage deviation calculated as; (HTP_S – HTP_D)/HTP_D, where HTP_S and HTP_D represent the posterior estimates of HTP from the sensitivity analysis and the default prior analysis respectively. The model was implemented in WinBUGS Version 1.4.1 with the first 10,000 iterations discarded as burn-in and 50,000 iterations used for posterior inference. Convergence was assessed by visual inspection of the time series trace plots and autocorrelation plots and by running multiple (n=3) chains from different starting values. Figures were constructed using the “ggplots2” package in R.

3. Results

198 **3.1. Descriptive Statistics**

199 **3.1.1. Secondary dataset (2005); Formulation of priors**

200 In total, there were 20,323 test results available from the 2005 dataset. After removing non-
201 relevant results, 5,822 test results from 119 herds were available in the final dataset. The modal
202 value for the prior for HTP was 0.32. The 95% confidence intervals were 0 - 0.92. The beta
203 distribution was fitted with a mode of 0.32 and a 95th percentile of 0.92. The resulting
204 distribution had alpha and beta parameters of 1.18 and 1.25 and 10th, 50th and 90th
205 percentiles of 0.12, 0.48 and 0.86 respectively. Within infected herds, the CWHP was 0.151 with
206 a mode at 0.1, the resulting beta distribution used for the prior had alpha and beta parameters
207 of 2.37 and 13.31 and 10^{th}, 50^{th} and 90^{th} percentiles of 0.051, 0.136 and 0.272 respectively.

208 **3.1.2. Primary dataset (2013-14)**

209 Descriptive statistics are shown in Table 3. After removing error records, data were available
210 for 99,101 animals in 1,039 dairy herds. Average herd size was 95.4 animals, the majority of the
211 herds were located in Leinster (n=249) and Munster (n=719) provinces and these herds also
212 had the greatest average herd sizes (108.5 and 102.1 respectively). Four hundred and forty
213 eight herds (43.1%) had an apparent prevalence of 0, i.e. no animals testing positive. The
214 distribution of apparent prevalence for herds with 1 or more animals testing positive is shown
215 in Figure 1.

216 **3.2. Model outcomes**

217 The median posterior estimate for HTP (95% posterior probability interval) was 0.28 (0.23,
218 0.32). Across all herds, the median ATP was found to be 0.032 (0.009, 0.145), whilst within
219 infected herds, the median CWHP was 0.137 (0.033, 0.348). Figure 2 shows the probability
220 distribution for HTP, along with the distribution of the probability of infection for all of the
221 herds.

222 **3.3. Sensitivity analysis**

Overall, the model was reasonably robust to each of the priors used in the analysis. Varying the mode and upper 95th percentile of each prior by up to 50% in either direction resulted in posterior median estimates for the HTP of between 0.265 – 0.323, which were within the 95% posterior probability interval of the original estimate. The posterior distribution for HTP was most sensitive to the prior for CWHP and to the Se estimate for the ELISA. In both cases, the direction of the change of the posterior was counter to the direction of the change for the prior. The model appeared to be relatively insensitive to variation around the prior for HTP and varying this prior by up to 50% in either direction resulted in deviations of less than 0.1% in HTP. Increasing test specificity led to a decrease in the posterior HTP whereas the converse was noted when the specificity was reduced. However, even when the specificity estimate was increased by 50%, the posterior estimate remained very similar, increasing from 0.280 to 0.288.

4. Discussion

This study represents the first use of Bayesian methodology to estimate the true prevalence of paratuberculosis in Irish dairy herds. The posterior estimate of HTP of paratuberculosis among dairy herds enrolled in the national control programme was 0.23 - 0.34 with a 95% probability.

Care must be taken when comparing prevalence studies which may have been conducted on different populations using different tests evaluating different target conditions. Previous to this study, only one HTP estimate had been published for paratuberculosis in Ireland (Good et al., 2009). The posterior HTP estimate from the present study was higher than that reported in the 2009 study (0.206) (Good et al., 2009). However, the earlier study utilised frequentist methods to estimate the true prevalence of herds with at least one infectious (shedding) animal and was based on a serological test Se of 0.278-0.289. The Bayesian methodology used in the current study however, incorporated uncertainty and variability associated with the test Se by modelling this variable as a probability distribution, the target condition in the present study was “infected” rather than “infectious” and the mode of the distribution used to model test Se was 0.15 and 0.22 depending on the test used. Finally, the previous study was based on data

collected in 2005. In the presence of a decline in the number of dairy herds, an increase in herd sizes, and in the absence of a nationally co-ordinated control programme, it is likely that HTP may have increased in the intervening years, resulting in the increased estimate observed in the present study.

It is noteworthy that within the population of herds enrolled in the national Control Programme, the estimated overall HTP is significantly lower in comparison to that reported for other countries. Nielsen and Toft (2009) estimated that HTP across Europe was likely to be greater than 0.5 based on limited information available at that time. More recently, Pozzato et al. (2011) found that HTP was likely to be approximately 0.7 in two regions of Northern Italy whilst Verdugo et al. (2015) found a trend of decreasing HTP over a 3-year period in Denmark from 0.92 to 0.75. Finally, Lombard et al. (2013) estimated the HTP in US dairy herds to be approximately 0.91.

However, the results of the present study should be interpreted with some caution in the wider context of the disease in Ireland. The primary (2013-2014) dataset used for the current study was based on test results collected from herds enrolled in a voluntary control programme with an average herd size of 95 cows, whereas the national average dairy herd size in 2014 was around 60 cows (Central Statistics Office, 2015; DAFM, 2015). Furthermore, given that herdowners join the national control programme voluntarily, it is likely that herds enrolled within the control programme may differ from the wider population of dairy herds in Ireland. Herd owners may have enrolled in the belief that their herd is free from the disease, with the aim of demonstrating freedom of their herd through the control programme. In this case it might be expected that HTP among herds enrolled in the scheme may be lower than that in the general population. However at the time of this study, a herd classification system was not yet introduced for the scheme, meaning that the benefit for the herd owner when the herd tested negative was not attainable by the farmer in the short term. Conversely herd owners may have joined the scheme in the belief or knowledge that their herd was infected in order to take

advantage of tools developed for control of the disease in infected herds. We might expect this to increase the HTP in the study in relation to the national herd level prevalence.

The results of the sensitivity analysis (Table 4) suggest that the model was reasonably robust to the selection of priors. Varying the priors by up to 50% had only a modest effect on the primary outcome of interest. Overall, the model was most sensitive to the prior for CWHP and diagnostic test Se.

Whilst conducting this research, a previously reported method for modelling CWHP was considered (Branscum et al., 2004). This method utilised a combination of a beta distribution and gamma distribution in order to model CWHP with the form; Beta($\mu\psi$, $\psi(1-\mu)$) where μ is a beta distribution and ψ is a gamma distribution. However, in attempting to use this method in the present study, we noted that the low CWHP and high degree of between-herd variability frequently pushed the parameters of this prior less than 1. The resulting beta distribution became increasingly clustered at 0 when increased variability was introduced. We therefore concluded that this method would not be appropriate for the present study. A single beta distribution was used to model CWHP which combined uncertainty and variability associated with this variable.

5. Conclusion

Paratuberculosis test records from 99,101 animals in 1,039 herds between November 2013 and December 2014 were used to produce a Bayesian estimate of HTP in Irish dairy herds. The median posterior estimate for HTP (i.e. the probability of a randomly selected herd containing at least one truly positive animal), among dairy herds enrolled in the national Johne's Disease Control Programme, was 0.28 (95% posterior probability interval; 0.23, 0.34).

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302 **References**

303 Alinovi, C. A., Ward, M. P., Lin, T. L., Moore, G. E., & Wu, C. C., 2009. Real-time PCR, compared to
304 liquid and solid culture media and ELISA, for the detection of Mycobacterium avium ssp.
305 paratuberculosis. Vet Microbiol, 136, 177-179.

306 Aly, S.S., Anderson, R.J., Whitlock, R.H., Adaska, J.M., 2014. Sensitivity and specificity of two
307 enzyme-linked immunosorbent assays and a quantitative real-time polymerase chain reaction
308 for bovine paratuberculosis testing of a large dairy herd. Int J Appl Res Vet Med 12, 1-7.

309 Berkvens, D., Speybroeck, N., Praet, N., Adel, A., Lesaffre, E., 2006. Estimating disease prevalence
310 in a Bayesian framework using probabilistic constraints. Epidemiology 17, 145-153.

311 Branscum, A., Gardner, I., Johnson, W., 2004. Bayesian modeling of animal-and herd-level
312 prevalences. Prev Vet Med 66, 101-112.

313 Brenner, H., Gefeller, O., 1997. Variation of sensitivity, specificity, likelihood ratios and
314 predictive values with disease prevalence. Stat Med 16, 981-991.

315 Chiodini, R.J., Chamberlin, W.M., Sarosiek, J., McCallum, R.W., 2012. Crohn's disease and the
316 mycobacterioses: a quarter century later. Causation or simple association? Crit Rev Microbiol
317 38, 52-93.

318 Chun-Lung, S. 2010. BetaBuster programme, Department of Medicine and Epidemiology,
319 University of California, Davis.

320 Central Statistics Office (CSO). 2015. Selected livestock numbers in December.
321 http://www.cso.ie/multiquicktables/quickTables.aspx?id=aaa06. Accessed 11/12/2015.

322 DAFM, 2015. Animal Health Computer System. Department of Agriculture, Food and the Marine,
323 Dublin, Ireland.

Enøe, C., Georgiadis, M.P., Johnson, W.O., 2000. Estimation of sensitivity and specificity of diagnostic tests and disease prevalence when the true disease state is unknown. Prev Vet Med 45, 61-81.

Geraghty, T., Graham, D.A., Mullowney, P., More, S.J., 2014. A review of bovine Johne's disease control activities in 6 endemically infected countries. Prev Vet Med 116, 1-11.

Good, M., Clegg, T., Sheridan, H., Yearsely, D., O'Brien, T., Egan, J., Mullowney, P., 2009. Prevalence and distribution of paratuberculosis (Johne's disease) in cattle herds in Ireland. I Vet J 62, 597.

Greiner, M., Gardner, I.A., 2000. Epidemiologic issues in the validation of veterinary diagnostic tests. Prev Vet Med 45, 3-22.

Hanson, T., Johnson, W.O., Gardner, I.A., 2003. Hierarchical models for estimating herd prevalence and test accuracy in the absence of a gold standard. J Agr Biol Envir St 8, 223-239.

Jubb, T. F., Sergeant, E. S. G., Callinan, A. P. L., & Galvin, J. W., 2004. Estimate of the sensitivity of an ELISA used to detect Johne's disease in Victorian dairy cattle herds. Aus Vet J 82, 569-573.

Lombard, J., Gardner, I., Jafarzadeh, S., Fossler, C., Harris, B., Capsel, R., Wagner, B., Johnson, W., 2013. Herd-level prevalence of Mycobacterium avium subsp. paratuberculosis infection in United States dairy herds in 2007. Prev Vet Med 108, 234-238.

McKenna, S.L.B., Keefe, G.P., Barkema, H.W. and Sockett, D.C., 2005. Evaluation of three ELISAs for Mycobacterium avium subsp. paratuberculosis using tissue and fecal culture as comparison standards. Vet Microbiol, 110, 105-111.

Messam, L.McV., Branscum, A.J., Collins, M.T., Gardner, I.A., 2008. Frequentist and Bayesian approaches to prevalence estimation using examples from Johne's disease. Anim Health Res Rev 9, 1-23.

More, S., Doherty, M., Downey, L., McKenzie, K., Devitt, C., O'Flaherty, J., 2011. Animal Health Ireland: providing national leadership and coordination of non-regulatory animal health issues in Ireland. Revue Scientifique et Technique-OIE 30, 715-723.

Norton, S., Johnson, W.O., Jones, G. and Heuer, C., 2010. Evaluation of diagnostic tests for Johne's disease (Mycobacterium avium subspecies paratuberculosis) in New Zealand dairy cows. J Vet Diagn Invest, 22, 341-351.

Nielsen, S.S., Toft, N., 2008. Ante mortem diagnosis of paratuberculosis: a review of accuracies of ELISA, interferon-γ assay and faecal culture techniques. Vet Microbiol 129, 217-235.

Nielsen, S.S., Toft, N., 2009. A review of prevalences of paratuberculosis in farmed animals in Europe. Prev Vet Med 88, 1-14.

Nielsen, S.S., Toft, N., Okura, H., 2013. Dynamics of specific anti-Mycobacterium avium subsp. paratuberculosis antibody response through age. PLoS ONE, 8 (2013), p. e63009

Pozzato, N., Capello, K., Comin, A., Toft, N., Nielsen, S.S., Vicenzoni, G., Arrigoni, N., 2011. Prevalence of paratuberculosis infection in dairy cattle in Northern Italy. Prev Vet Med 102, 83-86.

Rogan, W.J., Gladen, B., 1978. Estimating prevalence from the results of a screening test. Am J Epidemiol 107, 71-76.

Sweeney, R.W., 2011. Pathogenesis of paratuberculosis. Vet Clin N Am: Food Anim Pract 27, 537-546.

van Weering, H., van Schaik, G., van der Meulen, A., Waal, M., Franken, P., van Maanen, K., 2007. Diagnostic performance of the Pourquier ELISA for detection of antibodies against Mycobacterium avium subspecies paratuberculosis in individual milk and bulk milk samples of dairy herds. Vet Microbiol 125, 49-58.

370 Verdugo, C., Toft, N., Nielsen, S.S., 2015. Within-and between-herd prevalence variation of
371 Mycobacterium avium subsp. paratuberculosis infection among control programme herds in
372 Denmark (2011–2013). Prev Vet Med 121, 282-287.

373 Youden, W. J., 1950. Index for rating diagnostic tests. Cancer, 3: 32–35

374