The True Seroprevalence of Enteropathogenic Yersinia in Pigs, a Bayesian Approach


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Abstract
Bayesian inference was used to estimate the true seroprevalence of enteropathogenic Yersinia in pigs in Finland. Sensitivity and specificity of the diagnostic test were also estimated. One-hundred-seventy-two pigs of different ages were sampled and analysed for antibodies against enteropathogenic Yersinia outer proteins by a commercially ELISA test. Posterior probability estimates for sensitivity and specificity of the ELISA were 69.9% and 84.3%, respectively. The posterior probability of the true seroprevalence of enteropathogenic Yersinia was 78.6% for all pigs. There was an age tendency with the highest seroprevalence values in fattening pigs and in sows.

Introduction
Yersiniosis is a food-borne disease in humans, mainly caused by Yersinia enterocolitica (EFSA and ECDC, European Centre for Disease Prevention and Control 2012). Y. enterocolitica infections have been associated with the consumption of pork products (Tauxe and others 1987; Fredriksson-Ahomaa and others 2006; Rosner and others 2010). Healthy pigs are often asymptomatic carriers of Yersinia; and are considered the major reservoir of this zoonotic agent (Fredriksson-Ahomaa and others 2001; Fredriksson-Ahomaa and others 2006).

Serum samples can be analysed for antibodies against Yersinia by different serological test (Nielsen and others 1996; von Altrock and others 2006). In general, serology is a diagnostic tool that can be used for monitoring Yersinia, and it is cheaper and less time-consuming than the bacteriological methods (Fredriksson-Ahomaa and others 2011).

Detection of antibodies could provide a good estimation of the prevalence of enteropathogenic Yersinia in pigs at farms, when taking into account the sensitivity and specificity of the diagnostic tests used. The true seroprevalence can be estimated from an apparent seroprevalence by using the Bayesian inference which allows the incorporation of prior information in addition to the data. As well, the Bayesian approach provides a reliable estimate of the sensitivity and specificity when there is no gold standard test. The objective of the present study was to estimate the true seroprevalence of Yersinia in pigs using a Bayesian approach based on a cross-sectional sampling, and to estimate the diagnostic test sensitivity and specificity. The results showed that the sensitivity and specificity of the ELISA test were lower than previously reported by the manufacturer; and, the posterior probability estimate of the true seroprevalence was 78.6% (95%PI 61.5 – 90.5), for all pigs.

Material and Methods
In this study, serum samples were collected and analysed for occurrence of antibodies against enteropathogenic Yersinia. Individual serum samples from weaning pigs (20 to up to 50 kg), fattening (50 kg or more), and sows were collected on the farms, as previously described by Virtanen et al. (Virtanen and others 2012). The serum samples were tested for the presence of Yersinia antibodies by using a commercially available ELISA kit (Pigtype Yopscreen, Labor Diagnostik, Leipzig, Germany).

The definitions of apparent seroprevalence (Ap), true seroprevalence (Tp), sensitivity (Se) and specificity (Sp) were considered as defined by Thrusfield (Thrusfield 2007). Independent beta prior distributions were used to take into account the uncertainty in the true seroprevalence, sensitivity, and specificity (Hanson and others 2003). The number of seropositive pigs (x) is conditional on the true seroprevalence, thus the model was: x ~binomial (Ap, n), where Ap = Tp*Se + (1-Tp)(1-Sp).

Prior beta distributions for the true seroprevalence were constructed based on a systematic review of the literature. Average pooled results of the apparent prevalence and its 95% confidence interval for each age group were calculated. Finally, they were used as inputs in the software Betabuster (downloaded from http://www.epi.ucdavis.edu/diagnosticstests/betabuster.
Information provided by the ELISA test's manufacturer validation report was used to estimate the prior distributions for sensitivity and specificity of the serological analyses. The sensitivity was estimated assuming a binomial model and uniform prior, where x out of n infected animals tested positive, thus beta (x+1, n-x+1). The specificity was similarly estimated. Models were constructed in WinBUGS 1.4.3. Inferences were based in 50000 iterations after a burn-in of 1000 iterations for convergence. Results from the marginal posterior distributions are summarized as the median and their probability intervals (PI).

**Results**
The estimated sensitivity of the ELISA was 69.9% (95% PI 61.9 - 77.7), and the specificity was 84.3% (95% PI 51.9 – 99.2). The posterior probability estimates of the true seroprevalence in each age group of the Finnish pig population are presented in the Figure.

A sensitivity analysis was conducted using different beta prior distributions for each model, and no significant differences were found between the posterior estimates, F-value (p>0.05).

**Discussion**
The commercial ELISA has been used to determine the seroprevalence of *Yersinia* in pigs (von Altrock and others 2006; Fredriksson-Ahomaa and others 2009; von Altrock and others 2011; Virtanen and others 2012) without questioning the accuracy characteristics reported by the manufacturer. The estimations obtained indicated that the commercial ELISA had lower sensitivity and specificity than previously reported by the manufacturer.

Several studies have reported apparent seroprevalence of *Yersinia* in pigs; however, differences in sensitivity and specificity between diagnostic methods result in different true seroprevalence estimations. The true seroprevalence estimated in this study was significantly higher than the commonly reported apparent seroprevalence. The differences might be explained because those studies were based on a frequentist approach without taking into account the prior information nor the uncertainty of the sensitivity and specificity of the diagnostic test used. Therefore, the true seroprevalence estimated in the present study is not comparable directly.

Fattening pigs showed a true seroprevalence of 77.9%, value between the range of previously reported apparent seroprevalence of 2.5% (Nesbakken et al., 2007) to 82.1% (Virtanen et al., 2012). The use of two ELISA tests was reported to be used, which might explain the wide range of seroprevalence. The two test were the anti-LPS ELISA for specifically detection of *Y. enterocolitica* O:3 (Nesbakken and others 2006; Nesbakken and others 2007) and the same ELISA kit that we used that is against antibodies of enteropathogenic *Yersinia* (von Altrock and others 2006; von Altrock and others 2011; Virtanen and others 2012).

The lowest value of the true seroprevalence was found in weaning pigs, as they might be still protected by the maternal antibodies against *Yersinia*. Significant differences in seroprevalence between groups were observed, showing that the true seroprevalence increased with age, as previously reported by Vilar (Vilar and others 2013).

**Conclusion**
The Bayesian approach provided reliable information on the seroprevalence of enteropathogenic *Yersinia* in pigs, and also useful information of the ELISA diagnostic test commonly used to detect antibodies against *Yersinia*.
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