P21

Identification of potential risk factors for Toxoplasma gondii in fattening pigs in the Netherlands using a Bayesian approach

Eppink D.M.[1], Bouwknegt M.[1], Oorburg D.[1], Urlings H.A.P.[1], van Asseldonk M.A.P.M.[2], Van Wagenberg C.P.A.[2], Krijger I.[3], van der Giessen J.W.P.[4], Swanenburg M.[4], Wisselink H.J.[5]

[1]Vion Food Group, Boxtel, Netherlands, [2]Wageningen Economic Research, Wageningen, Netherlands, [3]Wageningen Livestock Research, Wageningen, Netherlands, [4]National Institute for Public Health and the Environment, Bilthoven, Netherlands, [5]Wageningen Bioveterinary Research, Wageningen, Netherlands

Introduction

Toxoplasma gondii is a relevant foodborne pathogen, it is estimated that up to one third of the world population has been exposed to the parasite (Tenter et al. 2000). In the Netherlands toxoplasmosis ranks second on a list of prioritized emerging zoonosis (Havelaar et al. 2010) and also second in disease burden among 14 foodborne diseases (Mangen et al. 2017). Data suggest that ingesting improperly cooked meat containing *T. gondii* is one of the major sources of infection in Europe and North America (Crotta et al. 2017; Guo et al. 2015). The contribution of pork to meatborne *T. gondii* infections is estimated to be 11% in the Netherlands (Opsteegh 2011) and is seen as an important possible source of human *T. gondii* infections (Foroutan et al. 2019). The European Food Safety Authority (EFSA) advised to perform serological testing of pigs and on farm audits on risk factors (EFSA 2011). To that end, a serological monitoring program was developed in a slaughterhouse in the Netherlands. In this study, the objective is to determine the association between within-herd seroprevalence, corrected for misclassification of samples through Bayesian analyses, and risk factors for *T. gondii* on fattening pig farms in The Netherlands.

Materials and Methods

From 2015 to 2018, HACCP based audits were performed on 75 fattening pig farms in The Netherlands to identify the presence of potential *T. gondii* risk factors. All farms were conventional pig farms, with 15 farms being farrow to finish. As overall seroprevalence of *T. gondii* in pigs in the Netherlands is low, estimated at 5% (1-12% 95% CI) by Foroutan et al. 2019, approached farms were chosen with the knowledge of previous serology data. In this way there would be farms with positive serum samples and farms without them included in the study. The audits were based on an updated version of the questionnaire from Mul et al. (2015) and covered the following topics: outdoor access, farm biosecurity, rodent control, presence of cats, feed and water supply. In addition, serum samples (n=6272) from fattening pigs were obtained at slaughter throughout the year before the audit on the farm was performed. These samples were used for antibody testing by a PrioCHECK™ Toxoplasma Antibody ELISA. Data were analysed using Bayesian statistics, with the within-farm *T. gondii* prevalence as dependent variable and potential risk factors as independent variables. As always with serology, misclassification due to false-positive or false-negative results can occur. Statistical methods have been developed to account for such misclassification, based on frequentistic as well as Bayesian approaches (Hui & Walter 1980; Joseph et al. 1995). First, all independent variables were analysed in a univariate logistic model, and variables with a probability ≤0.25 that zero is included in the 95% interval were analysed in a multivariable model. The multivariate logistic model was fitted using backward elimination until all remaining variables showed a probability ≤0.05 that zero is included in the 95% interval. Two-way interaction terms were evaluated similarly to the main variables regarding statistical significance.

Results

Descriptive results showed that 50 out of the 75 farms had 1 or more positive serum sample in the year before the audit was performed. In total 438 samples were positive out of the 6272 samples. Final Bayesian analyses are currently being conducted. However, preliminary results from data analysis using frequentistic logistic multivariate regression identified two significant risk factors: the accessibility of pig feed for cats and the provision of well water as drinking water for the pigs (Table 1).

Discussion and Conclusions

The use of serological testing seems to be a valuable guide and monitoring tool for the control of *T. gondii* in pork production. In a preliminary analysis, a higher within-herd *T. gondii* seroprevalence on fattening pig farms in the Netherlands was associated with the accessibility of pig feed for cats and the provision of well water as drinking water for the pigs. Improvements in farm management on fattening pig farms will likely contribute to reduction of the human disease burden and is presently studied.

References

Crotta M, Limon G, Blake DP, Guitian J. Knowledge gaps in host-parasite interaction preclude accurate assessment of meat-borne exposure to *Toxoplasma gondii*. International Journal of Food Microbiology 261 (2017) 95–101.

European Food Safety Authority; Scientific Report on Technical specifications on harmonized epidemiological indicators for public health hazards to be covered by meat inspection of swine. EFSA Journal 2011; 9(10):2371.

Foroutan M, Fakhric Y, Riahid SM, Ebrahimpoure S, Namroodif S, Taghipourb A, Spotingh A, Gamblei HR, Rostamie A. The global seroprevalence of *Toxoplasma gondii* in pigs: A systematic review and meta-analysis. Veterinary Parasitology 269 (2019) 42–52.

Guo, M, Dubey, JP, Hill, D, Buchanan, RL, Gamble, HR, Jones, JL, Pradhan, AK. Prevalence and risk factors for *Toxoplasma gondii* infection in meat animals and meat products destined for human consumption. J. Food Prot. 78 (2015), 457–476.

Havelaar, AH, van Rosse, F, Bucura, C, Toetenel, MA, Haagsma, JA, Kurowicka, D, Heesterbeek, JAP, Speybroeck, N, Langelaar, MFM, van der Giessen, JWB, Cooke, RM, Braks, MAB. Prioritizing emerging zoonoses in the Netherlands. 2010. PLoS ONE 5, e13965

Hui SL, Walter SD. Estimating the error rates of diagnostic tests. Biometrics 1980; 36: 167–171.

Joseph L, Gyorkos TW, Coupal L. Bayesian estimation of disease prevalence and the parameters of diagnostic tests in the absence of a gold standard. American Journal of Epidemiology (1995); 141: 263–272.

Mangen, MJ, Friesema, IHM, Pijnacker, R, Mughini Gras, L, Van Pelt, W (2018). Disease burden of food-related pathogens in the Netherlands 2017; Bilthoven; National Institute for Public Health and the Environment.

Opsteegh M. *Toxoplasma gondii* in animal reservoirs and the environment. Dissertation Utrecht University 2011, Faculty of Veterinary Medicine

Tenter, AMAR, Heckeroth, and LM Weiss. *Toxoplasma gondii*: from animals to humans. Int. J. Parasitology 2000; 30: 1217–1258.

Table 1: Variables analysed multivariably by backward elimination for association with the presence of T. gondii on 75 Dutch pig farms between 2015 and 2018 (univariable P ≤ 0.25)

Risk Factor	N Farms	Odds Ratio (95% CI)	P-Value
Goats Absent Present	 67 8	Not applicable	0.176
Boots in stable Only inside Also outside	 28 47	Not applicable	0.524
Professional pest control Yes No	 33 42	Not applicable	0.283
Own cats at barnyard Absent Present	 42 33	Not applicable	0.850
Pigfeed accessible for cats Absent Present	 49 26	15.4 (3.0 – 79.4)	0.001
Pig drinking water Tap water Well	 34 41	3.4 (1.1 – 10.7)	0.035
Pigfeed contains whey Absent Present	 52 23	Not applicable	0.429
Pigfeed Dry feed Wet/liquid feed	 37 38	Not applicable	0.069