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Research paper

Short- and long-term association between individual levels of milk antibody against *Ostertagia ostertagi* and first-lactation heifer's production performances

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ABSTRACT

It is agreed that exposure of adult dairy cattle to helminths on pasture can negatively affect production performances as milking herd. Young animals, especially replacement heifers, represent the future of a dairy farm and are among the most vulnerable to helminth infections in a dairy herd. For this reason, dairy farmers tend to frequently treat heifers against helminths, although the impact of helminths on heifers' production performances is still poorly understood. Using different epidemiological and serological tools, this study examines the relationship between heifer exposure to helminths on pasture and production performances over time. During a one-year period, 1,454 individual milk samples were collected from first-lactation heifers in England and tested for *Ostertagia ostertagi* (*O. ostertagi*) antibodies. After controlling for other confounders, increasing milk antibody levels against *O. ostertagi* were significantly associated with decreased milk yield at sampling but not at day 305 of heifer lactation. We did not observe any relationship between milk antibody levels against *O. ostertagi* in heifers and yields in fat and protein. However, heifers with a high level of milk antibodies against *O. ostertagi* were more likely to produce dead calf at first calving and present a delay in second calving. Moreover, these heifers had significantly higher levels of milk antibodies against *Mycobacterium paratuberculosis* (*M. paratuberculosis*) during their first lactation and were more likely to die before the end of the study. We argue that epidemiological approaches can be useful but must be complemented by other methodologies to better understand the impact of helminth infections in dairy heifers. In order to address the complex dynamics of helminth infections in dairy cattle production we require more comprehensive approaches that include triangulation between data sources and interdisciplinary studies.

1. Introduction

Worldwide, there is an increasing demand for food, especially meat and milk (FAO, 2009). Alongside this demand, and due to growing concerns around food production sustainability (i.e. the need for increased food production with less waste and environmental impact) and other issues such as animal welfare, high expectations are put on livestock systems not only for increasing production and efficiency but also for complying with sustainability and ethical standards (Rushton and Bruce, 2016). According to recent reports, the global production of animal-source food is reduced by 20% due to diseases (Vallat, 2009). Cattle helminth infections represent one of the growing concerns for the cattle industry around the world (Skuce et al., 2013). Intensification of cattle production as well as changes in climate and management practices have affected the distribution of helminth infections in cattle (Rushton and Bruce, 2016). In fact, in recent years, the incidence of chronic diseases due to cattle helminth infections has increased along with evidence of parasite resistance to cattle anthelmintic drugs (Stafford and Coles, 1999; Pritchard et al., 2005; Skuce et al., 2013).

In temperate areas such as England, there is a general agreement that cattle helminths, particularly *Ostertagia ostertagi* (*O. ostertagi*), are of major importance in terms of their economic impact on the dairy livestock system (Skuce et al., 2013; Charlier et al., 2014; Sargison, 2014). However, to date there is no systematic and agreed approach to assess the costs associated with cattle helminth infections (Rushton and Bruce, 2016). In this context, there is a need for better understanding the biological processes underlying cattle helminth infections, in particular *O. ostertagi*, under real farm conditions.

A number of studies have been conducted on farms to understand the effects of helminth infections on cattle milk production and

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reproductive performances (Sanchez et al., 2004a). Some of these studies have shown that effective treatments for subclinical helminth infections are associated with increasing milk production (Sanchez et al., 2004a; Charlier et al., 2007b; Verschave et al., 2014). A meta-analysis of published literature estimated that, after controlling for study bias, anthelmintic treatments were associated with a daily milk increase of 0.35 kg/cow/day (Sanchez et al., 2004a). However, such an approach does not take into account the effect of different helminths and exposure levels on production losses. In addition, evidence suggests that anthelmintic drugs could directly stimulate cow milk production (Purvis and Whittier, 1996). In other studies, high levels of bulk tank milk antibody against *O. ostertagi* were associated with an annual drop of cow milk production (Sanchez and Dohoo, 2002; Charlier et al., 2005). However, the use of pooled samples also makes the interpretation of these results difficult (Sekiya et al., 2013). In addition to these effects on milk production, cattle helminths could also reduce calving interval and number of breeding at conception and increase the mortality rate in a dairy herd (Walsh et al., 1995; Stromberg et al., 1997; Sanchez et al., 2002a; Delafosse, 2013). Interestingly, although heifers represent a capital investment for dairy farmers and are among the most vulnerable to this type of infections and production losses, little has been done to explore impacts of helminth infections in first-lactation heifers, with very few, inconclusive studies available (Blanco-Penedo et al., 2012; Liedtke et al., 2013). Moreover, it is not clear whether losses in milk yield due to helminth infections can be compensated during the subsequent lactations of the cow. Finally, although there is clear evidence that *O. ostertagi* actively suppresses cattle immune responses (Gasbarre, 1997), there is no evidence from studies conducted on farms of the effects of this parasite on cattle susceptibility to other diseases.

Climatic conditions and herd management vary greatly between countries, which ultimately influences measures of impact (Williams, 1999; Sanchez et al., 2002a). Moreover, infections such as helminth infections affect cattle systems at different levels (e.g. animal, farm, livestock sector and national) and dimensions (e.g. milk production, reproduction, health and welfare), for which the individual level represent a start (Rushton and Bruce, 2016). In this study, we examine the relationship between individual exposure to helminths on pasture and the production performances of first-lactation heifers in England, taking the gastrointestinal nematode (GIN) *O. ostertagi* as a case study. Besides overcoming methodological limitations in the current literature, we also discuss the value of epidemiological approaches in assessing the effects of cattle helminth infections on production performances under real farm conditions.

2. Materials and methods

2.1. Study heifers

Since individual milk (IM) antibody levels against *O. ostertagi* highly vary within-farm (Charlier et al., 2007a), the sampling aimed to sample more heifers per farm across the seasons than farms. Heifers came from a convenience and purposive sample of dairy farms, all members of the Quality Milk Management Services (QMMS) recording scheme, Somerset, England. Farms were selected to allow the representation of different levels of heifer exposure to helminths on pasture and heifer management. Farm inclusion criteria included heifers calving all-year-round or at least during two different seasons in a year, home rearing of heifers (i.e. not contract reared), compliance with data recording, agreeing with the study protocol and sharing farm records. There were no restrictions on the type of cattle housing (i.e. housed all-year-round, in the winter only, and varied) or the practices of anthelmintic treatments. In total, 43 English dairy farms were included in the study. The average size of herds sampled was 150 cows, of which 46 were first lactation heifers. Heifer IM samples were obtained from samples routinely collected and stored by QMMS. The determination of dairy heifer sample size involved both statistical and non-statistical considerations (e.g. time, budget, and farm recording). These were aligned to the study objectives of identifying significant association between outcomes (i.e. heifer production, reproduction and health) and predictors (i.e. *O. ostertagi* milk antibodies) (Dohoo et al., 2009). Heifer sample size calculation was based on available estimates of association between anti-*O. ostertagi* milk antibody levels and milk production in adult cows (Sanchez et al., 2004a). Considering the origin of the farms, no estimate of likely dropouts and withdrawals was taken into consideration in the heifer sample size determination. A total of 1500 heifers were included in the study from March 2014 to March 2015 - with 35 heifers (i.e. 1,500/43) regularly sampled throughout the seasons on each farm and tested for *O. ostertagi* antibodies. A more detailed description of the heifer samples selection criteria and the sampling process is available in Bellet et al. (2018).

2.2. Data collection

Detailed retrospective and prospective information on demographic and management was obtained for each heifer, from birth to the end of the study (i.e. one year after the last heifer sampling). These included information on housing, food (including grazing), vaccination and anthelmintic treatments, before and after individual sampling. The collection of data relied on the use of different tools and approaches, including questionnaires, face-to-face and telephone interviews and QMMS' information management system. Individual parameters of heifers' milk production, reproduction and health were extracted from QMMS laboratory's information management system and processed using the dairy herd data analysis program TotalVet (QMMS Ltd/SUM-IT Computer Systems). In order to collect one-year of prospective production data for each heifer, data covered the period between March 2014 (i.e. start of the milk samples collection) and April 2016 (i.e. the end of the study). At the time of milk sampling (t_S), heifers' individual records included season, age, breed, milk yield, fat, protein, somatic cell counts (SCC), calving date and status of offspring (i.e. alive or dead). Cumulative milk, protein and fat yields of heifers at day 305 of heifer lactation (t_{305}) were obtained if heifers had reached this stage at the end of the study (t_E). These were calculated beforehand by QMMS, using the 'test-interval' method (ICAR, 2016). The interval between the first and second calving of heifers was computed from the corresponding calving dates, if present. Since farmers' assiduousness to record varied by farm and variables, only accurate health variables with a sufficient number of observations were extracted from TotalVet and considered for the analysis. These health variables included individual levels of milk antibody against *Mycobacterium paratuberculosis (M. paratuberculosis)* during the first lactation of heifers and heifer's health status at t_E (i.e. present, dead and absent (culled or dead)).

2.3. ELISA milk testing

Considering the fact that heifer samples would be stored for a period of several months before testing, a pilot study was conducted to evaluate the effect of milk sample storage for over a one-year period on ELISA results using IM samples from adult cows. Cow samples that had been tested for *O. ostertagi* antibodies in 2012 and then stored at QMMS at −20 °C, were tested again under similar laboratory conditions in March 2014. No significant differences were obtained between the results of the two years (Bellet et al., 2018). After collection on farm, heifer IM samples were preserved using bronopol/natamycin and kept at ambient temperature until arrival at the laboratory. In the laboratory, the samples were processed, tested for SCC, fat and protein, before being frozen at −20 °C (± 2 °C) until further testing; this was achieved within the first 48 h after samples collection on farms. In order to account for possible cross-reactivity between the *O. ostertagi* test and *Fasciola hepatica* (*F. hepatica*) (Bennema et al., 2009), levels of farm exposure to *F. hepatica* were determined by antibody-detection ELISA