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# Climate-driven longitudinal trends in pasture-borne helminth infections of dairy cattle

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## ABSTRACT

Helminth parasites of grazing ruminants are highly prevalent globally and impact negatively on animal productivity and food security. There is a growing concern that climate change increases helminth disease frequency and intensity. In Europe, these concerns stem from case reports and theoretical life cycle models assessing the effects of climate change scenarios on helminth epidemiology. We believe this study is the first to investigate climate-driven trends in helminth infections of cattle on a cohort of randomly selected farms. One thousand, six hundred and eighty dairy farms were monitored over an 8 year period for the two major helminth infections in temperate climate regions and climate-driven trends were investigated by multivariable linear mixed models. The general levels of exposure to *Fasciola hepatica* decreased over the study period while those to *Ostertagia ostertagi* increased, and this could at least be partially explained by meteorological factors (i.e. the number of rainy (precipitation >1 mm) and warm days (average daily temperature >10 °C) in a year). The longitudinal trends varied according to the altitude and the agricultural region of the farm. This study shows that longitudinal epidemiological data from sentinel farms combined with meteorological datasets can significantly contribute to understanding the effects of climate on infectious disease dynamics. When local environmental conditions are taken into account, the effects of climate change on disease dynamics can also be understood at more local scales. We recommend setting up a longitudinal sampling strategy across Europe in order to monitor climate-driven changes in helminth disease risk to inform adaptation strategies to promote animal health and productivity.

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## 1. Introduction

Worldwide, helminth parasites of grazing ruminants impact negatively on animal productivity and food security (Fitzpatrick, 2013). In dairy cattle, infections with *Ostertagia ostertagi* and *Fasciola hepatica* are among the most significant helminth infections and are responsible for major economic losses annually (Charlier et al., 2014). The annual cost per adult cow in Flanders, Belgium was estimated at €38 and €30 for *O. ostertagi* and *F. hepatica*, respectively (Charlier et al., 2009).

Increases in helminth-associated disease frequency and intensity have been reported within the European ruminant sector in recent years (van Dijk et al., 2010). Given the strong dependence of the life cycle of helminths on meteorological conditions, the role of climatic factors has been put forward to explain this observed

resurgence of helminth-associated disease, and the field of forecasting parasite risk based on these conditions is attracting renewed attention (Mas-Coma et al., 2008). Two main approaches have been employed: (i) a normative approach where knowledge of the life cycle is translated into a mathematical model. This model is subsequently fed by observed meteorological conditions or climate change scenarios to forecast helminth disease risk for the short or long term (e.g. Rose et al., 2016) and (ii) an empirical approach where indices of parasite infection are measured in the field and correlated with climate-environmental variables to predict disease risk at un-sampled locations (e.g. Ducheyne et al., 2015). These correlative models, however, are typically cross-sectional and do not allow identification of drivers of longitudinal trends in infection risk (Austin, 2007). These limitations are further complicated by the fact that if longitudinal data are available, those are mostly opportunistic, passive surveillance data, whereas in order to build reliable models, purpose-driven, active surveillance data are needed (Fox et al., 2012).

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Here, we report the results of an empirical cohort study of 1,680 randomly selected dairy farms that were monitored over an 8 year period for the two major helminth infections in a temperate climate region. We use statistical models to assess the long-term trend in exposure to infection as well as the role of meteorology and environment to explain variations in exposure between farms and years. To our knowledge this is the first large cohort study to investigate climate-driven trends in helminth infection in ruminants.

## 2. Materials and methods

### 2.1. Study area

The study was conducted in Flanders which has a surface area of 13,587 km<sup>2</sup> and a temperate maritime climate. The average annual temperature and precipitation in Belgium over the period 1981–2010 were, respectively, measured as 10.5 °C and 852 mm from 199 rainy days ([www.meteo.be](http://www.meteo.be)). Elevation in Flanders varies slightly, from sea level in the west to 150 m above sea level in the south and east. Large areas of the total surface (24.3%) are prone to flooding (Van Orshoven, 2001). At the start of the study (2006), 7002 dairy farms in Flanders provided milk to a dairy cooperative. The average number of dairy cows per herd was 48 (Bernaerts et al., 2008) and the majority of the herds (98%) had pasture at their disposal with the average grazing season being 6.5 months (Bennema et al., 2010).

### 2.2. Selection of farms and sample collection

Selection of the farms was previously described by Bennema et al. (2009). In total, 1,800 farms were selected from a list containing all dairy herds in Flanders that provided milk to a dairy cooperative ( $N=7,002$ ). One thousand, four hundred farms were randomly selected, stratified at the community level. Additionally, 400 herds were selected randomly from the remaining herds in the list. This procedure was chosen to ensure that the whole study area was covered in the sampling and that 25% of the total study population was included in the study. The locations of the sampled farms and study regions are shown in Fig. 1.

Bulk-tank milk samples were obtained from the selected herds in the months of October or November from 2006 until 2013. Not all of the selected farms could be sampled each year due to logistical reasons. In some cases, the sample volume was too small and it was only possible to test for one parasite. Furthermore, due to farms ceasing their professional activities, there was a gradual decline in the number of sampled herds over the study period. The numbers of data available for analysis are reported in Table 1. In some cases, two bulk-tank milk samples originating from the same farm were collected in the same year. In this case, both samples were analysed and the average result of the two measurements was used.

### 2.3. Bulk-tank milk ELISAs for the detection of antibodies against *F. hepatica* and *O. ostertagi*

The level of exposure to *F. hepatica* and *O. ostertagi* was determined by antibody-detection ELISAs applied on bulk-tank milk and test results were expressed as an optical density ratio (ODR), which is the measured optical density (OD) of a test sample corrected for the OD of a negative and positive control that were run on each plate.

The ELISA for *F. hepatica* was performed as described by Charlier et al. (2007a) and uses excretory–secretory products of *F. hepatica* as antigen. From 2012 onwards, an in-house ELISA (Charlier et al.,

2007a) was replaced by a commercially available assay based on the same antigen (SVANOVIR® *F. hepatica*-AB ELISA, Boehringer Ingelheim Svanova, Uppsala, Sweden). A pairwise comparison of both tests revealed a strong correlation ( $R^2 = 0.89–0.94$ ) but consistent lower ODRs for the SVANOVIR® assay. Therefore, the results of the commercial ELISA were transformed according to the formula  $ODR_{transformed} = ODR_{original} * 1.250 + 0.048$  in order to enable comparison with the results from previous years (2006–2011).

Antibodies against *O. ostertagi* were quantified using the SVANOVIR® *O. ostertagi*-AB ELISA (Boehringer Ingelheim Svanova), following the instructions of the manufacturer. Both ELISAs have previously been used as indicators of parasite exposure as well as parasite-induced production losses (Charlier et al., 2014).

### 2.4. Meteorological and environmental variables

A geographic information system database was constructed containing environmental as well as meteorological data that were considered to have a relevant impact on the epidemiology of *F. hepatica* and *O. ostertagi* infections in cattle (Bennema et al., 2011). Sampled herds were first geo-referenced by farm address and then the nearby pasture was geo-referenced on the centroid using the data layer of Flemish agricultural properties.

Environmental data layers included were the Digital Elevation Model (DEM) and the hydrological atlas of Flanders. From the DEM, slope and elevation were calculated. From the hydrological atlas, distance to water and areas prone to flooding (yes/no) were derived. Agricultural region (a categorical variable with seven levels) was used as a proxy for soil type. The different agricultural regions are shown in Fig. 1.

Daily temperature and precipitation data from 2006 to 2013 were obtained from the Royal Meteorological Institute of Belgium (RMI) for 20 weather stations. Meteorological data for the farms was obtained using an inverse distance weighted interpolation based on the six nearest weather stations. Based on the daily measurements, the following variables were calculated: number of rainy days (precipitation >1 mm), annual precipitation (mm), number of warm days (average daily temperature >10 °C) and average annual temperature (°C). The temperature of 10 °C was chosen as this is a critical threshold above which the development of *F. hepatica* in the environment and intermediate host snails can take place (Torgerson and Claxton, 1999). While low rates of development of the eggs and pre-infective stages of *O. ostertagi* have been observed from 4 to 6 °C onwards, consistent development is also observed at temperatures >10 °C (Pandey, 1972; Young et al., 1980). The meteorological variables show variations both between and within farms over time. Therefore, each of these variables was split into ‘between’ and ‘within’ farm components. The ‘between’ component is the farm-specific average measurement of the variable over the years. The ‘within’ component is the deviation of the measurement of the variable in each year from the overall farm average. This approach allows differentiation of the ‘space’ from ‘time’ effects of a variable.

### 2.5. Data analysis

The purpose of the analysis was to investigate changes in the levels of exposure to parasite infections over time and to see whether those changes could be explained by the selected meteorological and environmental variables. We used general linear mixed-effects models for continuous longitudinal data (Verbeke and Molenberghs, 2000), which take the form:

$$Y_i = X_i\beta + Z_i b_i + \varepsilon_i$$

where  $Y_i$  is the  $n_i$ -dimensional response vector for farm  $i = 1, \dots, N$ .  $X_i$  and  $Z_i$  are  $(n_i \times p)$  and  $(n_i \times q)$  known design matrices, respectively.

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