# Parasitic infections in an organic grazing cattle herd in Tuscany using geographic information systems to determine risk factors

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#### Summary

An organic grazing cattle herd in Tuscany (Italy) was monitored for parasites between 2002 and 2006. Every two to three months, faecal samples from cattle of different breeds and age were collected and examined for endoparasites, using both qualitative and quantitative parasitological techniques. Several environmental parameters were monitored and data on biodiversity and field margin biodiversity of grazing areas were also collected. All data were geo-referenced and plotted on a vectorial map using geographic information systems (GIS) software. Soil was classified as silt and clay/sand. The hydraulic drainage was poor and water pooling was observed frequently. The biodiversity of field margins was relatively high. Cattle were bv infected coccidia, gastrointestinal nematodes, cestodes and trematodes. Prevalence and intensity of infestations were highly variable. In most cases, this variability was related to cattle breed, age, season and meteorological data. The Pisana breed was most commonly infected by Fasciola hepatica and Paramphistomidae. These infestations were associated with more frequent flooding and water pooling in the areas grazed by this breed.

#### Keywords

Cattle, Climate, Endoparasites, Environment, Geographic information system, Grazing, Italy, Risk, Tuscany.

# Endoparassitosi bovine in una azienda biologica toscana: utilizzo del sistema informativo geografico per la valutazione dei fattori di rischio

#### Riassunto

Nel periodo di tempo compreso tra il 2002 ed il 2006, una azienda biologica di bovini da carne della Toscana è stata monitorata per le endoparassitosi connesse con il pascolo. A tal fine, campioni fecali di bovini di razza ed età diversa sono stati raccolti ogni due-tre mesi ed esaminati per la ricerca di endoparassiti utilizzando tecniche parassitologiche sia quali che quantitative. Sono stati anche monitorati numersosi parametri ambientali e raccolti dati riguardanti la biodiversità vegetale e la biodiversità dei margini dei pascoli. Tutti i dati sono stati georeferenziati e disegnati su una mappa vettoriale utilizzando il software GIS. Il terreno è stato classificato come fangoso e argilloso/sabbioso. Il drenaggio idraulico è risultato insufficiente e la formazione di ristagni idrici è stata osservata frequentemente. La biodiversità dei margini dei

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campi è risultata relativamente elevata. I bovini sono risultati infetti da coccidi, nematodi gastrointestinali, cestodi e trematodi. La prevalenza e l'intensità di queste diverse endoparassitosi è stata molto variabile ed in molti casi correlata alla razza, all'età, alla stagione ed ai dati meteorologici. La razza pisana è risultata più frequentemente infetta da Fasciola hepatica e Paramfistomidi e ciò è stato messo in relazione alle inondazioni ed al ristagno delle acque più frequenti osservati nelle aree pascolate da questa razza.

#### Parole chiave

Bovini, Endoparassiti, Pascolo, Rischio, Sistema informativo geografico, Toscana.

## Introduction

The development of methods for mapping diseases has progressed considerably in recent years. Geographic information systems (GIS) offer a new tool for epidemiological studies. GIS has been applied to study the spatial and temporal patterns of parasitic infestations (6, 9, 16).

Organic animal production relies heavily on grazing in pastures where soil-transmitted parasitic infections can be prevalent. These economically important diseases have negative effects on animal health, welfare and production (5, 7, 21). In organic cattle herds, the control of parasitic diseases is difficult because of grazing management and limitations on the amount and frequency of drug use (11, 19). For these reasons, continuous parasitological monitoring is essential for the efficacious control of parasitic diseases (17, 17). This data is most useful when parameters, linked with environmental including climatic data and soil and land characteristics. To obtain useful data for a large-scale parasite control programme of ruminants organically reared in Tuscany, an organic cattle herd was selected for a pilot study to monitor endoparasitic diseases and their spatial and temporal distribution.

# Materials and methods

In this study, the *La Sterpaia* organic cattle herd in the Natural Park of Migliarino-San RossoreMassaciuccoli (Pisa) was monitored from May 2002 to March 2006 to:

- evaluate the presence, prevalence and intensity of endoparasitic diseases and their association with age and breed of cattle, farm management, environmental parameters and season
- identify and evaluate the risks associated with endoparasitic diseases.

## La Sterpaia organic farm

The farm and its grazing areas are part of San Rossore Park (Pisa), located around *La Sterpaia* buildings near the Serchio River in the northern part of San Rossore Park (1.607.400°E, 4.843.780 N, Gauss-Boaga projection).

The farm is composed of nine pasture units, each of 8-9 ha. The different cattle breeds present on the farm, namely: Limousine, Pezzata Rossa and Pisana in 2002-2003 and Chianina, Pisana and Limousine in 2004-2006 are allowed to graze in different areas (Figs 1 and 2). Plant species in these areas are both grazed species and a large number of ungrazed species. Grazing is integrated with supplemental feed during certain periods of the year.

## Parasitological techniques

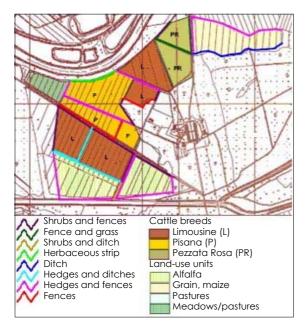
Between 2002 and 2006, about 300 cattle were examined every two to three months. Individual and pooled faecal samples were collected from naturally infected cattle. A description of the examination of samples is given below.

## Qualitative methods

The following methods were used:

- flotation test, using a low specific gravity solution (sg: 1 200) for *Coccidia*, gastrointestinal nematodes and cestodes (2, 8, 12, 14)
- flotation test using a high specific gravity solution (sg: 1 450) for *Dicrocelium dendriticum* (2, 8)
- Baermann method for lung strongyles (2, 8)
- sedimentation for *Fasciola hepatica* and Paramphistomidae (2, 8, 12).
- faecal cultures for infective stage (L3) gastrointestinal strongyle larvae (8, 10) to identify the percentage of infestation of genera (8, 12, 22)

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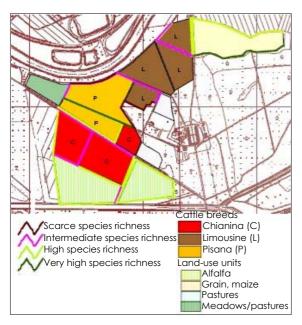


#### Figure 1

Distribution of cattle breeds in different grazing areas and field boundary structure on land units, 2002-2003

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed



#### Figure 2

Distribution of cattle breeds in different grazing areas and field boundary structures on land units, 2004-2006

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed • faecal cultures for *Eimeria* oocysts to identify species (8, 14).

The percentage of positive samples indicated the prevalence of each parasitic disease.

### Quantitative methods

A modified McMaster method (2, 15) was used to estimate the faecal egg count (FEC) for gastrointestinal nematodes (eggs per gram of faeces [EPG]) and *Eimeria* spp. (oocysts per gram of faeces [OPG]). The FEC indicated the intensity of coccidial and gastrointestinal nematode infections.

## Statistical analysis

EPG and OPG data were examined using analysis of variance (ANOVA). Associations with date of sampling, age and breed were tested. Data on strongyles and coccidia were transformed logarithmically [y = log(X + 25)] to normalise variance (8).

## **Environmental analysis**

Several environmental parameters, such as meteorological data (1) (Tables I and II), soil physical and chemical characteristics (3, 4), hydraulic and drainage parameters (4, 18) and land cover and land use were monitored. In addition, data on biodiversity and field margins (4, 13) of grazing areas were collected and classified using principal components analysis (PCA) using Canoco 4.5 (20), based on the boundary's structural attributes recorded in the field (shrub and tree cover, maximum and average height of vegetation, width and depth of ditches and the presence of a fence or herbaceous field margin).

Land-use heterogeneity (H) was determined using the following formula:  $H = -\Sigma p(i,j) * \log p(i,j)$  where p(i,j) is the proportion of the total number of pixels and land use types *i* and *j* are adjacent. Seven different land-use types were defined, namely:

- sown annual crops
- alfalfa
- pasture
- natural woodland
- poplar plantation
- rivers
- constructions.

Six transects of 2.25 km were divided into 10 pixels each and H was calculated based on land-use sequences between the pixels in these transects. Maximum land-use heterogeneity in the six transects was 1.45.

All data were geo-referenced (Garmin<sup>™</sup> global positioning system [GPS]) and drawn on a 1:10 000 digi-watcher proprietary video format (DGW) vector map (*Carta Tecnica Regionale* [CTR], Regional Agency for the Agricultural

#### Table I

Monthly mean temperature, rainfall and relative dampness in San Rossore Park, March 2002-May 2003

Date	Mean temperature (°C)	Rainfall (mm)	Relative dampness
Mar 2002	11.2	2.2	71%
Apr 2002	13.3	49	75%
May 2002	17.3	8.4	79%
Jun 2002	21.7	56.8	78%
July 2002	22.2	32	79%
Aug 2002	21.6	149.8	84%
Sept 2002	18.2	219.4	81%
Oct 2002	15.4	87.4	87%
Nov 2002	13.4	148	86%
Dec 2002	8.8	110.8	86%
Jan 2003	6.1	10.8	81%
Feb 2003	5.2	12.8	65%
Mar 2003	10.2	14.8	76%
Apr 2003	14	70.8	72%
May 2003	22	1.4	77%

Source: Agenzia Regionale per lo Svilippo e l'Innovazione nel Settore Agricolo-forestale-Regione Toscana, Servizio Agrometeorologico Regionale

#### Table II

Monthly mean temperature, rainfall and relative dampness in San Rossore Park, October 2004-March 2006

Date	Mean temperature (°C)	Rainfall (mm)	Relative dampness
Oct 2004	17.1	141.4	89%
Nov 2004	10.4	108.3	86%
Dec 2004	7.9	102.0	85%
Jan 2005	5.3	40.9	88%
Feb 2005	4.3	49.1	76%
Mar 2005	10.2	43.2	80%
Apr 2005	12.8	51.9	74%
May 2005	18.1	43.5	78%
June 2005	21.9	29.6	78%
July 2005	23.3	10.0	70%
Aug 2005	22.4	33.8	79%
Sept 2005	19.1	200.2	81%
Oct 2005	17.3	95.3	87%
Nov 2005	13.4	150	86%
Dec 2005	8.6	110	84%
Jan 2006	6.2	35.1	81%
Feb 2006	7.3	42.7	70%
Mar 2006	11.4	50.6	80%

Source: Agenzia Regionale per lo Svilippo e l'Innovazione nel Settore Agricolo-forestale-Regione Toscana, Servizio Agrometeorologico Regionale

Development and Innovation), using GIS software (ArcGIS<sup>TM</sup>). In addition, data on the different grazing areas were also processed by creating vector maps.

# Results

Grazing areas were classified as silt and clay/sand soil, with a mean percentage of organic material around 2%. The hydraulic drainage was poor; thus many ditches in the drainage system were occluded by dead vegetation. For these reasons, water pooling was frequently observed in some areas, especially during rainy periods. The biodiversity of field margins was relatively high (Figs 1 and 2), but land use for natural and cultivated pastures for animal nutrition reduced the landscape heterogeneity. Landuse heterogeneity was 1.12, 77% of Hmax; the matrix surrounding cultivated areas - which consists of planted poplar wood, pine wood, semi-natural woodland and a river contributed most to land-use heterogeneity in the study area. Within the cultivated area, little natural vegetation was observed. The main part of the Serchio area is used as meadows or pasture and consequently the soil is covered all year, with low land-use diversity related to the cultivated area only.

Eight boundary structure classes were identified by ordination analysis, based on the structural characteristics of the boundaries recorded in the field (Table III). The boundary structure was not correlated with land use of the adjacent fields because of the wide variety of boundary structure types and the relatively homogeneous land-use types (mainly meadows and pastures in the Serchio area). Most boundaries were characterised by the presence of a fence and this was possibly due to the presence of animals in all of the fields at some point in the crop rotation.

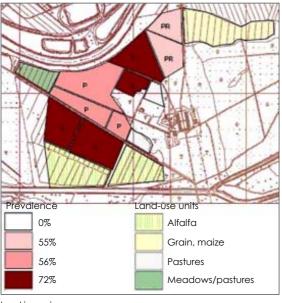
Regarding floral diversity, 127 plant taxa (between genus and species) have been recorded in the Serchio area. Species richness varied from 11 and 48 species per boundary. The average species richness per boundary structure class was variable but the structurally simpler boundaries belonging mostly to classes 4 and 8 had the lowest plant species richness (Table III). Distribution of the boundary classes was evenly spread over the entire area, indicating good spatial heterogeneity of the boundaries (Fig. 1). No correlation between plant species richness and type of disturbance (grazing, cutting) or land use was found (Fig. 2).

During all study periods, several species of parasites were identified. However, prevalence and, in the case of coccidia and gastrointestinal strongyles, intensity were highly variable. In most cases, this variability was associated with breed (Figs 3, 4, 5, 6, 7 and 8), age, season and meteorological data. In the first period examined (May 2002-May 2003), coccidiosis (mean prevalence 74%) was highly prevalent, with decreasing intensity during the year. Gastrointestinal strongyles (mean prevalence 96.7%) showed a higher prevalence and intensity in autumn and spring, while Strongyloides papillosus (mean prevalence 1.6%) and Neoascaris vitulorum (mean prevalence 3.2%) were found only in May 2002 and 2003. The highest prevalence of Moniezia spp. (mean prevalence 11.4%) was found in October 2002 and *F. hepatica* (mean prevalence 28.6%) between January and May 2003. D. dendriticum prevalence (mean prevalence 31.3%) increased between July 2002 and March 2003. During the first two years of monitoring, the Pisana breed was the only breed infected by F. hepatica Paramphistomidae (Fig. 7) and (mean prevalence 5.8%), with higher prevalence in bulls; infestation was associated with more frequent flooding and water pooling observed in the area used for the grazing of this cattle breed (Figs 1 and 2). The frequently observed water pooling is most probably due to the absence of ditches in the surrounding field boundary elements (Table III; Figs 1, 7 and 8). Decreased prevalence of coccidia (over than 90% in unweaned calves) and gastrointestinal helminths (gastrointestinal strongyles, S. papillosus, N. vitulorum and Moniezia spp.) were associated with increased age and the increase of liver fluke infestation was also observed in older animals. The former (coccidia and gastrointestinal helminths) were considered an important cause of production

#### Table III

Partitioning of the eight boundary structure classes identified in the Serchio area and their average plant species richness

Class	Boundary structure description	Boundaries (%)	Average plant species richness(n°)
1	Boundary with shrubs and a fence	23.5	29
2	Large herbaceous margin next to a fence	14.7	31
3	Boundary with shrubs and at least one ditch	2.9	29
4	Herbaceous field margin	2.9	13
5	Ditch with or without fence	2.9	48
6	Hedge with at least one ditch	8.8	29
7	Hedge with a fence	35.3	35
8	Small herbaceous margin under a fence	8.8	26



L Limousine

P PisanaPR Pezzata Rossa

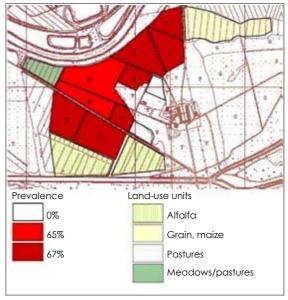
#### Figure 3

Mean prevalence of Coccidia (*Eimeria* spp.) in the different cattle breeds in an organic beef herd, 2002-2003

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed

losses in young animals and the latter (flukes) in adult animals. Between 2004 and 2006, breed (Chianina, Limousine and Pisana) was associated with gastrointestinal strongyles EPG and coccidian OPG. Growth phase (cow, unweaned calf, weaned calf and bull) was associated with OPG (p=0.006), while EPG was associated with season (p=0.046). The prevalence of flukes (mean prevalence 28.6%,



- C Chianina
- L Limousine
- P Pisana

#### Figure 4

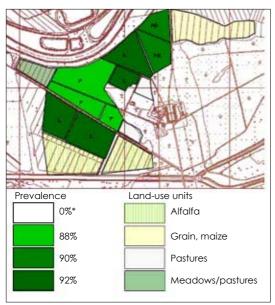
Mean prevalence of coccidia (*Eimeria* spp.) in the different cattle breeds in an organic beef herd, 2004-2006

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14% and 9.5% respectively for *F. hepatica*, *D. dendriticum* and Paramphistomidae) increased between the autumn of 2005 and spring of 2006 and was higher in Pisana and in cows of all breeds. The prevalence of coccidian (mean prevalence 67.4%) was higher in Limousine, especially in unweaned calves (90%). The prevalence of gastrointestinal strongyles (mean prevalence 77.5%) was

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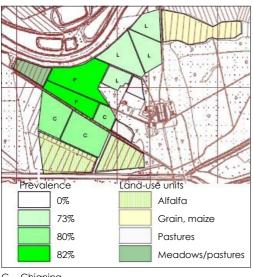
- L Limousine
- P Pisana PR Pezzata Rossa

#### Figure 5

Mean prevalence of gastrointestinal strongyles for the different cattle breeds in an organic beef herd, 2002-2003

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed



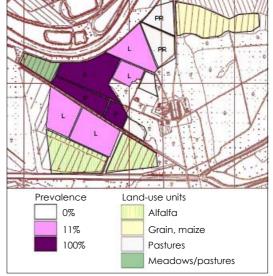
- C Chianina L Limousine
- P Pisana

#### Figure 6

Mean prevalence of gastrointestinal strongyles for the different cattle breeds in an organic beef herd, 2004-2006

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed



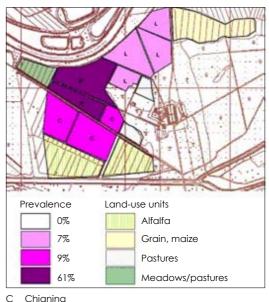
- L Limousine
- P Pisana
- PR Pezzata Rossa

#### Figure 7

Mean prevalence of Fasciola hepatica for the different cattle breeds in an organic beef herd, 2002-2003

Grazing areas are identified by the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed



- L Limousine
- P Pisana

#### Figure 8

Mean prevalence of Fasciola hepatica for the different cattle breeds in an organic beef herd, 2004-2006

Grazing areas are identified with the capital letter of each breed

Other areas (land use units) represent areas to use, in turn, as pastures in subsequent years or as supplemental feed

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higher in unweaned calves (>80%) of all breeds. The prevalence of S. papillosus (mean prevalence 11.2%) was highest in Limousine cows. N. vitulorum (mean prevalence 5.6%), gastrointestinal strongyles and S. papillosus showed higher prevalence and intensity of infestations in autumn. The prevalence and intensity of coccidiosis increased during the period under study. Eimeria bovis and E. zurnii were predominant in unweaned calves of all breeds, while E. alabamensis and E. ellipsoidalis occurred only in Limousine cattle. The gastrointestinal strongyles present in the herd were as follows: Ostertagia spp., Haemonchus spp., Cooperia spp., Trichostrongylus spp., **Bunostomum** Nematodirus spp., spp., Oesophagostomum spp. The greatest variability was recorded in the Chianina breed. The highest prevalence of Moniezia spp. (mean prevalence 4.5%) was found in March 2006, particularly in Chianina.

# Discussion

Times and methods of sampling and the number of samples collected were determined by several factors related to the management system used on the study farm. Sampling was only possible when livestock, normally grazing in different pastures and dispersed from one another, were moved and contained for other management reasons (for example, vaccination and treatment). Thus, some sampling bias was present. In addition, there were changes in some farm management practices during the last period of the study, possibly resulting in higher rates of contact among the different breeds. Despite these deficiencies, the data obtained showed a clear picture of the parasitological problems present in the farm.

As reported in previous studies (21), prevalence, intensity and number of endoparasites isolated in the La Sterpaia farm demonstrate the importance of endoparasitic diseases in organic farms. Coccidia (Eimeria spp.), nematodes (gastrointestinal strongyles, S. papillosus N. vitulorum), and flukes (F. hepatica, D. dendriticum, Paramphistomidi) and cestodes (Moniezia expansa and *M. benedeni*) were identified. The absence of the lung-worm (*Dictyocaulus viviparous*) during all study periods (2002-2006) confirms previous observations of its absence from this area (2).

Among the endoparasitic diseases detected, coccidiosis and gastrointestinal strongylosis were the most prevalent. Highly pathogenic coccidian species, such as E. bovis, E. zuernii and E. alabamensis, and the most pathogenic genera of gastrointestinal strongyles (2, 19) were isolated. Higher prevalence and infection loads for these two parasitic diseases were observed in younger animals and in warmer months (spring and autumn). For example, acute clinical coccidiosis was observed in May 2006 in Chianina and Limousine calves, at the same time as high OPG counts. In the case of gastrointestinal strongyles, qualitative and quantitative data obtained in this study showed high breeding risks (2) in all breeds, mainly in the final period of the study. Although the prevalence and intensity of S. papillosus, N. vitulorum and Moniezia spp. infestations were generally low, a high risk (especially for *N. vitulorum*) was observed: the prevalence in bulls was similar to that found in calves and young stock (2).

The prevalence of flukes was higher in winter and in older cattle, representing an important health problem and a cause of reduced production. During the first two years of monitoring, F. hepatica (Figs 7 and 8) and Paramphistomidae infestation was only detected in the Pisana breed; infestation was associated with frequent flooding and water pooling, mostly due to the absence of ditches in the surrounding field boundary elements observed in the area used for the grazing of this cattle breed. However, in the final part of the study, infestation in the two other breeds present on the farm (Chianina and Limousine breeds) was also observed (although at a lower prevalence). This infestation might have been linked to the higher contact rates between cattle of the different breeds during this period, probably enabling the spread of these two parasites through all breeds on the farm. During the first few months of 2006, several cases of mortality in Chianina cows and young

stock due to heavy *F. hepatica* infestations were observed on the farm. The absence of clinical infestations in the Pisana cattle, despite higher prevalence of *F. hepatica* infestation, could indicate a higher resistance of this breed to the pathogenic effects of *F. hepatica*.

# Conclusion

Results obtained in this study show that in the organic *La Sterpaia* cattle herd, prevalence and

intensity of endoparasitic diseases, and the health and breeding risks linked thereto, were associated with factors such as age and breed and with several environmental and management factors (grazing area, season and meteorological data, field boundary structures and hydraulic and soil drainage variables).

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