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Genetic factors of sheep affecting gastrointestinal parasite infections in the Distrito Federal, Brazil

Concepta McManus [a,*], Helder Louvandini [a], Samuel Rezende Paiva [b], Amaury Apolonio de Oliveira [c], Hymerson Costa Azevedo [c], Cristiano Barros de Melo [a]

[a] Faculdade de Agronomia e Medicina Veterinária, Universidade de Brasília, Brasília, Distrito Federal, CP 04508, 70910-900 Brazil
[b] EMBRAPA, Recursos Genéticos e Biotecnologia, Parque Estação Biológica - PqEB, Av. W5 Norte (final), Caixa Postal 02372, Cep: 70770-900, Brasília, DF, Brazil
[c] EMBRAPA, Centro de Pesquisa Agropecuária dos Tabuleiros Costeiros, Av. Beira Mar, 3250 - 13 de Julho, Caixa Postal 44, Cep: 49025-040, Aracaju, SE, Brazil

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A B S T R A C T

Three sheep farms were used in the Distrito Federal, Central Brazil, to study the occurrence of parasites in the feces. A total of 1798 collections were taken over the period of a year. A total of 1205 were taken in Santa Inês breed (SI) in all three farms, 323 in Bergamasca (Berg), 54 in Ile de France, 49 in Ile × SI, 103 in Morada Nova (MN) and 64 in Texel × SI, these last five groups being on a single farm. The animals were drenched soon after weaning and feces collected every 3 weeks to calculate fecal egg count (FEC), at least on two occasions on each animal. In some cases, blood was collected to determine packed cell volume (PCV) at fecal collection. Fixed effects included farm, breed/genetic group within farm, animal age (months), birth type (simple, twin) and sex. (Co)variance components were estimated for Santa Inês sheep using restricted maximum likelihood under an animal model. FECs were affected by month and farm showing that climate and management are important sources of variation for the parasites studied. While age and birth type of the lambs did not affect infection level, their genetic group was important, showing that breeding strategies can help control these parasites. Heritabilities for infection level in the sheep varied between 0.09 for *Strongyloides* and 0.31 for *Moniezia expansa*. Genetic selection strategies for sheep aimed at reducing these infections should result in more resistant animals.

1. Introduction

Gastrointestinal parasites are a major health problem in sheep flocks in Brazil, with *Haemonchus contortus*, *Cooperia* sp., *Trichostrongylus* sp., *Moniezia expansa* and *Oesophagostomun* sp. being the most commonly found species (Cenci et al., 2007). High stocking rates at pasture, associated with high environmental temperatures and regular rainfall in tropical conditions, mean high production per hectare but this leads to high infection rates for gastrointestinal parasites (Sotomaior and Thomaz-Soccol, 2001). According to Amarante and Barbosa (1995) the population dynamics of gastrointestinal helminthes and the interference of related environmental effects such as season, temperature and humidity have been important study points for the control of worms in ruminants. Bianchin and Melo (1985) showed that helminth larvae, in native or cultivated pastures, have their survival and maintenance controlled by climatic conditions, with higher contamination at the beginning of the rainy season and lower contamination in periods of lower precipitation.

Yamamoto et al. (2004) did not observe significant differences in larvae behaviour infecting pastures during

* Corresponding author. Tel.: +55 61 3307 2805/48; fax: +55 61 32736593.
E-mail address: concepta@unb.br (C. McManus).

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summer and winter periods in São Paulo State, Brazil. Nevertheless, there was a decreasing linear response in larvae number on the grass species in the upper third of plants in function of excessive sun exposure. In addition, Almeida et al. (2005) observed that environmental conditions allowed for the development of nematode larvae on the inside of the fecal bolus and their survival for long periods of time, represent a source of pasture contamination.

According to Siqueira (1993), farming in smaller areas, with permanent grazing and high stocking rates, favours the increase of helminth infection. Borba et al. (1993) confirm that, in a sheep herd, less than 5% of the parasite populations are found in the gastrointestinal tract of the animals, the rest are found on the pasture.

The capacity of sheep to acquire and express resistance to helminthes varies between breeds and individuals within breeds (Stear and Murray, 1994). Natural resistance to helminth infection in certain sheep breeds was found by Borba et al. (1997) and Amarante et al. (1999) who observed that native breeds (Crioula Lanada and Florida Native) were less susceptible to infection than specialized breeds (Corriedale and Rambouillet, respectively). In Brazil, the Santa Inês breed has been claimed to be resistant to mixed gastrointestinal parasite infections (Moraes and Thomaz-Soccol, 2001; Bueno et al., 2002; Amarante et al., 2004; Rocha et al., 2004). The objective of this study was to evaluate environmental and genetic factors affecting the level of gastrointestinal parasite infections (*Strongyloides*, Strongylodea, *Moniezia* and *Eimeria* sp.), as well as estimate genetic parameters for infection levels, in sheep in the Distrito Federal, Center-West of Brazil.

2. Materials and methods

The climate in the region is classified as AW according to the Köppen climatic classification system, with a mean annual temperature of 21.1 °C, with 16 °C and 34 °C as absolute minimum and maximum, respectively. Mean annual precipitation is 1578.5 mm^3 and mean relative air humidity is 68%. The climate is characterized by two very distinct seasons, a wet (summer) season where almost all of the precipitation occurs (October to April) and the other dry (winter), almost without rain (May to September).

A total of 1798 individual feces collections were carried out in three farms in the Distrito Federal, over the period of 1 year. These three farms are within 40 km of each other, two in Rural Nucleus PAD-DF on a red latosol at 1200 m above sea level and one in Rural Nucleus Vargem Bonita, on a red-yellow latosol at 1000 m. The farms had frequently exchanged rams and ewes in their 20-year history and in addition Santa Inês (SI) rams, half-brothers, were lent to each farm to create additional genetic links between them. On farm 3 there were also two Bergamasca, two Ile de France, two Texel and three Morada Nova rams. Of the animals sampled, 1205 were SI, 323 Bergamasca (Berg), 54 Ile de France (IF), 49 Ile × SI, 103 Morada Nova (MN) and 64 Texel × SI. The SI lambs were offspring of 657 ewes, with 234 lambing twice within the period. These sheep are not seasonal breeders.

The animals were weaned at 3 months of age, dewormed and feces were collected 3 weeks later, and a second time after 3 more weeks. In farm 3 blood was taken by venopunction from some animals (143) to detect packed cell volume (PCV) at fecal collection (Table 2). Animals were kept in *Andropogon gayanus* pasture, with a mineral salt *ad libitum* and corn silage during the dry season. Stocking rate varied from 4 AU/ha in the rainy season to 1 AU/ha in the dry season, with farm 3 tending to have higher stocking rates (approx 25%+) and all farms used a rotational grazing system. No lambs were castrated.

2.1. Parasitological exam

To monitor the fecal egg count (FEC) the modified Whitlock methodology (Ueno and Gonçalves, 1998) was used. The genera monitored were the nematodes *Strongyloides* and Strongylida, taenia *M. expansa* and fecal oocyte count (FOC) was monitored for the protozoa *Eimeria*. PCV was determined using microhaematocrit with centrifugation.

2.2. Statistical analysis

Data were analysed using the GLM Procedure of SAS® to identify the main sources of variation. Parasite occurrence was transformed by $\log_{10}(x+10)$. Factors evaluated included farm, breed/genetic group within farm, animal age, birth type (single or twin) and sex. Principal component analyses were also carried out using PRINCOMP procedure.

(Co)variance components for the parasite level in the SI breed were estimated using restricted maximum likelihood under an animal model. Heritabilities, permanent environment, as well as genetic and environmental correlations were estimated by MTDFREML (Multiple Trait Derivate Free Restricted Maximum Likelihood) (Boldman et al., 1995) using alterations by Kachman and Van Vleck (2007) and adjusting FEC:

$$Y = X\beta + Z_1 a + e$$

in which Y is a $(N \times 1)$ vector of observations on the animal; β vector of fixed effects (month and farm), associated with the incidence matrix X; a, vector of direct genetic effects associated with the incidence matrix Z_1; and e, vector of random residuals.

3. Results

The summary of variance analysis is in Table 1 with adjusted means in Table 2. PCV in the animal was affected by month and breed, while FEC and FOC were both affected by month, farm and breed, except for *M. expansa* which was not affected by breed.

Santa Inês sheep had a lower PCV than Ile × SI, both measures were within normal ranges (McManus et al., 2009). Morada Nova and Bergamasca had lower FEC for Strongylida, while lowest values for *Strongyloides* were found in Santa Inês and its cross with Ile de France (Ile × SI). The lowest FOC was found in Ile de France sheep. Lowest PCV was found at the beginning (March) and

Table 1
Summary of analysis of variance for packed cell volume (PCV), fecal eggs (FEC) and oocysts count (FOC) per gram in sheep feces in the Distrito Federal, Brazil.

	PCV (%)	Strongylida (FEC)	*Strongyloides* (FEC)	*Moniezia expansa* (FEC)	*Eimeria* sp. (FOC)
R^2	0.24	0.22	0.22	0.08	0.13
CV	19.43	211.64	103.09	454.43	640.74
Mean	32.03	286.85	0.70	14.89	31.31
Farm	–	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$
Month (farm)	$P < 0.0001$	$P < 0.0001$	$P < 0.0001$	$P = 0.0055$	$P < 0.0001$
Breed	$P = 0.0123$	$P = 0.0253$	$P = 0.0047$	ns	$P = 0.0347$
Birth type	ns	ns	ns	ns	ns
Sex	ns	ns	ns	ns	ns
Age	ns	ns	ns	ns	ns

ns = not significant, CV: coefficient of variation, and R^2: coefficient of determination.

Table 2
Mean of packed cell volume (PCV), fecal eggs (FEC) and oocysts count (FOC) per gram in lambs in the Distrito Federal per breed, sex, birth type and month.

	PCV (%)	Strongylida (FEC)	*Strongyloides* (FEC)	*Moniezia expansa* (FEC)	*Eimeria* sp. (FOC)
Breed					
SI[a]	35.91a	347.88c	19.14a	35.33	0.62b
MN[b]		8.65a			
Bergamasca		193.99b	49.49b	31.29	0.66b
Ile		484.62c	29.57ab	53.57	0.18a
Ile × SI	42.75b	482.26c	17.72a	35.70	0.29ab
Texel × SI		279.50bc	39.02b	2.07	0.94b
Sex					
M	36.17	247.36	15.08	31.09	0.61
F	42.48	235.92	1.51	30.43	0.46
Birth type					
1	38.09	171.25	6.48	24.66	0.58
2	40.56	312.04	22.99	36.86	0.49
Month					
J	39.1bc	190.81b	3.03a	22.18a	0.33a
F		237.96bc	8.19a	21.79a	0.36a
M	36.99a	137.64b	4.38a	22.35a	0.72b
A		154.89b	6.83a	24.32a	0.59b
M	38.39ab	23.56a	28.63b	12.80a	0.76b
J		429.15c	20.76ab	43.59ab	0.58b
J	34.79a	214.35b	37.55b	4.44a	0.38a
A		225.97b	0.91a	32.00ab	0.28a
S		310.88bc	10.94a	94.00c	0.16a
O	43.32c	1306.14d	7.81a	30.90a	0.52ab
N	43.32c	8.94a	8.51a	43.81ab	1.39c
D		41.07a	4.61a	25.82a	0.37a

Means in the same column followed by the same letter do not differ significantly by the Tukey test ($P > 0.05$).
[a] Santa Inês.
[b] Morada Nova.

middle (July) of the dry season, associated with higher Strongylida infections. Infections were highest in the middle of the dry season for *Strongyloides* and start of the wet season for Strongylida (Table 2). *M. expansa* showed a peak at the end of the dry season (September) and *Eimeria* sp. levels were highest at the start of the dry season, with another peak at the beginning of the rainy season.

Genetic correlations between the FEC or FOC and parasites species were low (Table 3). The highest (negative) correlation was between PCV and Strongylida (−0.32), while the correlation between PCV and *Eimeria* sp.

Table 3
Genetic correlations (with standard errors) and heritabilities (h_d^2) between fecal egg counts and packed cell volume in Santa Inês sheep in the Distrito Federal, Brazil.

	Packed cell volume	Strongylida	*Strongyloides*	*Moniezia expansa*	*Eimeria* sp.
Strongylida	−0.32 (0.07)				
Strongyloides	0.15 (0.05)	0.21 (0.07)			
M. expansa	0.15 (0.06)	0.04 (0.04)	−0.02 (0.03)		
Eimeria sp.	0.31 (0.05)	0.04 (0.04)	0.02 (0.02)	0.01 (0.03)	
h_d^2	0.15 (0.09)	0.18 (0.05)	0.09 (0.06)	0.31 (0.10)	0.24 (0.09)

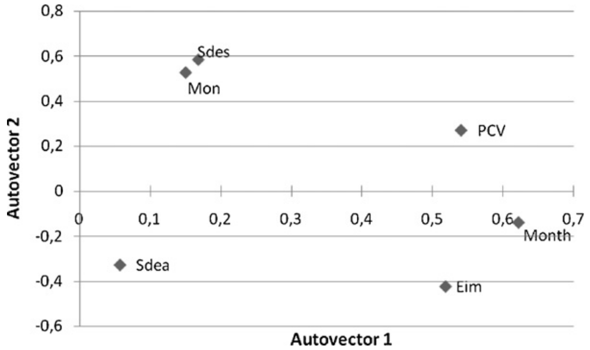

Fig. 1. First two autovectors for parasite infection in lambs in the Distrito Federal, Brazil. PCV: packed cell volume; Sdes: *Strongyloides* sp.; Mon: *Moniezia*; Eim: *Eimeria* sp.; Sdea: Strongylida.

was 0.31. When Strongylida infection increased, PCV decreased, indicating greater anemia in animals with higher infection. Heritabilities varied from 0.09 to 0.31 and standard errors were in general high compared to the estimates.

Principal component analysis is in Fig. 1. The first two autovectors explain 49% of all the variation between the traits. An increase in *Eimeria* sp. infection was accompanied by an increase in PCV. For the second autovector, an increase in *Strongyloides* and *M. expansa* infection was accompanied by an increase in PCV and a decrease in Strongylida and *Eimeria* sp.

4. Discussion

Although Santa Inês are frequently marketed as being resistant to endoparasites, this was not confirmed here. This may be due to the recent history of this breed which is today the largest sheep breed in Brazil. Studies have shown that the Santa Inês has two distinct genetic groups (Paiva et al., 2005), one which is the result of crossbreeding to improve carcass quality, probably with Suffolk sheep, and then selected for the lack of residual wool. McManus et al. (2009) found no significant differences between black and brown Santa Inês (derived from the above mentioned cross) or crossbred Bergamasca with Santa Inês in terms of heat tolerance. Bricarello et al. (2005) also found no significant differences between Santa Inês and Ile de France lambs under mild *H. contortus* infection for haematological and biochemical profiles. This may be also due to poor productivity of the Santa Inês, as it has a lower growth rate than other terminal sire breeds. This lower genetic potential for growth affects resilience, as less demand is put on nutritional partition of factors which affect both growth and resilience such as protein level in the feed (Bricarello et al., 2005; Louvandini et al., 2006).

Fecal egg count (FEC) has been used as an indicator of resistance in sheep (McEwan et al., 1992; Amarante et al., 1998). Breeds with European blood (pure or crossbred) showed higher infection levels than hair breeds (SI and MN). These breeds may be naturally more resistant to parasite infections. This is in agreement with Amarante et al. (2004), who showed that while 100% of SI lambs were resistant to infection, 80% Suffolk lambs were susceptible, under the same management conditions. Bueno et al. (2002) observed that SI animals were less susceptible to nematode infection and better adapted to intensive management conditions compared to Suffolk, Ile de France and Poll breeds.

Nieto et al. (2003) did not observe significant differences ($P > 0.05$) in FEC carried out in lambs from crosses of Corriedale ewes with Bergamasca and Hampshire Down rams, but month had a significant effect over FEC ($P < 0.05$). Breed was also found to be an important source of variation by Marley et al. (2003), showing the possibility of selection and substitution of animals to control the infection, but animal age did not affect infection level. Here there was little difference in age between the animals studied, so further studies are needed with animals of varying ages.

In the present study, a higher FEC was observed in the dry season. This confirms the observations by Braga and Girardi (1991), Bianchin and Melo (1985), and Honer and Bianchini (1987) who concluded that strategic control should concentrate in the season with greatest hydrodeficit, as this will reduce the number of helminthes in the host, and the environment will be unfavourable to free living forms, reducing pasture infestation.

Peak elimination of *Eimeria* sp. has great importance in the maintenance of environmental contamination and infection of different categories of sheep, as type of environment, nutritional and heat stress, as well as the ingestion of a large number of oocysts of *Eimeria bakuensis* was found to cause the clinical form of the illness (Foreyt, 1990). Nevertheless, Hassum et al. (2002) observed that animals eliminated *Eimeira* sp. oocysts during all months of the year, and with reduction in peaks probably associated with intercurrent disease. Taylor and Catchpole (1994) related extreme temperatures, diet changes and other adverse conditions to reduction in resistance to coccidia. It was seen that in the transition period between the end of the rainy season and start of the dry season, there was an increase in *Eimeria* oocysts. This time is marked by a reduction in the quantity and quality of pasture available to the animals, but supplementation with concentrate has still not been put in place. This may affect host resilience through nutritional factors (Louvandini et al., 2006).

Moniezia sp. was present in all months of the year, with an increase in the end of the dry season (September). This is in agreement with Braga and Girardi (1991) who observed *Moniezia* sp. in tracer animals on native pasture in Roraima State, Brazil and concluded that precipitation was the most important climatic factor for determining development and survival of larvae in the environment. Nevertheless, Kawano et al. (2001) observed *M. expansa* in all lambs up to 120 days of age, but with different infection levels.

Although there were no significant differences between sex and birth type, in general lambs born as twins had slightly higher levels than those born as singles. This is probably due to competition for colostrum which offers passive protection for new-borns (Baracat et al., 1997).

Forage species may also significantly affect the development and survival of infectant larvae of helminthes in the feces of the host animal (Marley et al., 2003). In all farms studied *Andropogon* sp. was the dominant pasture type, by differences between management strategies and local environmental conditions may have affected the difference in levels of resistance between farms.

Heritabilities for infection level in the Santa Inês breed varied between 0.09 for *Strongyloides* and 0.31 for *M. expansa*. Literature estimates in other breeds and regions are in agreement with this observation. Bisset et al. (1992, 1994, 1996) found heritabilities in traits associated with FEC between 0.11 and 0.34 in Romney sheep in New Zealand and Douch et al. (1995) found a medium heritability (0.23) in the same breed for two sample dates. In Australia, Eady et al. (1996), working with different strains of Merino tested for resistance to endoparasites, estimated heritability between 0.07 and 0.42 for the different strains. Pollott et al. (2004) showed heritability for FEC with animals in the same breed to be 0.28 ± 0.072, varying from 0.21 to 0.60, increasing with the age of the animal. Other studies (Bouix et al., 1998 in Polish long-wool and Rege et al., 1996 in Ethiopia, for Menz and Horro breeds) found heritabilities varied between 0.20 and 0.34, depending on the collection month. Few resistance estimates are available in Brazil. Nieto et al. (2003) used a threshold model and estimated heritability as 0.08, lower than that here, but animals found to have higher FECs were dewormed, which may have affected this parameter.

5. Conclusion

The environment significantly affected FEC in lambs reared in the Distrito Federal, Brazil, so control strategies should take this into account. Genetic selection strategies for sheep aimed at reducing these infections should result in more resistant animals.

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