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Research paper

Faecal egg counts and immune markers in a line of Scottish Cashmere goats selected for resistance to gastrointestinal nematode parasite infection

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ABSTRACT

This study aimed to investigate the effect of selection for low faecal egg count (FEC) in Scottish Cashmere goats in comparison to a control line of unselected goats grazing the same pasture. Goats from generations F2 through to F9 were monitored for FEC, bodyweight, peripheral eosinophilia and IgG, IgA and IgE response against *Teladorsagia circumcincta* from the end of their first grazing season, through winter housing (during which a single artificial challenge dose of 10,000 drug susceptible *T. circumcincta* was given) and the following full grazing season. The study demonstrated that selected line animals excreted a significantly lower number of parasite eggs ($P < 0.01$) in the majority of generations examined. Liveweight productivity was unaffected by selection. Although selected line animals had greater numbers of circulating eosinophils in many of the generations (four generations of males and six generations of females, $P < 0.05$), there was no direct link between eosinophilia and reduced FEC. Immunoglobulin levels showed no consistent difference between selected and control lines. IgG, IgA and IgE levels were not different between lines over the whole dataset

($P > 0.05$), although the selected line had significantly elevated or reduced levels ($P < 0.05$) for all three within individual generations. There were significant associations between increased IgG and reduced FEC under artificial infection conditions ($P = 0.02$). Increased IgA was also significantly associated with elevated FEC during the second grazing season ($P < 0.001$). The study demonstrates that selection produced a line of goats with consistently reduced FEC compared with control animals, but did not identify a clear relationship between any of the immune markers measured and faecal egg output.

1. Introduction

With reliance upon anthelmintics for livestock production increasingly under threat due to widespread anthelmintic resistance, the development of alternative means of parasite control is paramount. One of the most promising means of reducing reliance upon chemical control is selective breeding for animals with increased responsiveness to infection (Gray, 1997; Stear et al., 2001; Gruner et al., 2002; Eady et al., 2003) due to the heritability of responsiveness (Gauly and Erhardt, 2001), repeatability of faecal egg counts (FEC) in individuals (Stear et al., 1995a, 1995b), and variation between animals (Stear and Murray, 1994). Responsiveness to gastrointestinal parasites is often measured as a repeatable reduction in faecal egg count as this is the most practicable method of measuring parasite infection level (Gray, 1997). A number of studies (Dawkins et al., 1989; Patterson et al., 1996a, 1996b; Windon, 1996; Woolaston and Piper, 1996) in both sheep and goats have shown that responsiveness is a highly heritable phenomenon, and that, in sheep, high numbers of peripheral eosinophils are a useful marker of responsiveness (Dawkins et al., 1989; Stear et al., 2002).

In goats, Patterson et al. (1996a, 1996b) found that individuals with low faecal egg output had correspondingly high numbers of peripheral and tissue eosinophils, in addition to lower worm burdens, increased retardation of worm development, and increased or no difference in the numbers of mucosal mast cells or globule leucocytes. Immunoglobulin has also been implicated in the immune response to nematodes; Stear et al. (1995a, 1995b) found a significant negative correlation between both plasma and mucosal IgA

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and faecal egg count, although no relationship between IgG_1 or IgG_2 and egg count was observed. Studies by Huntley et al. (2001) on lambs infected with *Teladorsagia circumcincta* have shown higher levels of IgE in the sera of lambs with lower faecal egg output, suggesting a protective role of IgE in the response to nematode infection. This was further confirmed by Pettit et al. (2005) who found higher numbers of IgE-bearing cells and Murphy et al. (2010) who observed higher IgE against both larval antigens and recombinant *T. circumcincta* in lambs with lower faecal egg counts.This study investigated the potential for selection of a line of Scottish Cashmere Goats with increased responsiveness to gastrointestinal parasites. Goats are implicated in accelerating the development of anthelmintic resistance (Varady et al., 2011) due to their increased susceptibility to infection (Le Jambre and Royal, 1976; Pomroy et al., 1986), differences in their pharmacokinetics (Sangster et al., 1991) and reduced immune response to gastrointestinal parasites (Huntley et al., 1995) compared with sheep. This has clear implications where goats are grazed with or in close proximity to sheep in terms of spreading resistant strains between host species (Varady et al., 1994).

In this study, data from eight successive generations of selectively bred goats co-grazed with unselected control animals was examined in an attempt to confirm previous studies on selection for responsiveness and to identify the mechanism behind the low egg output phenotype. This was achieved through examination of parasitological and productivity data and immunological and biochemical measurements performed on stored plasma samples.

2. Materials and methods

2.1. Animals

The goats used in the study originate from a group of animals initially imported in 1986 from Iceland, Tasmania, New Zealand and Siberia as part of a selection programme designed to breed a line of cashmere-producing goats (Scottish Cashmere Goats) suited to the Scottish climate at Sourhope Research Station in the Scottish Borders. A helminth selection line was established from these animals in 1992, initially using 95 2–4 year old breeding does randomly drawn from the wider population. Initial studies established the feasibility of selecting breeding does using low FEC by identifying 'responder' and 'non-responder' animals (Patterson et al., 1996b) and the selection of sires based on the repeatability of FEC (Patterson et al., 1996a) A line of goats selected for low FEC was established from these animals. A breeding herd of 95 does was established using randomly selected animals initially, but subsequently replaced with selected progeny when does become too old or infirm for further breeding.

2.2. Selected line breeding criteria

Trichostrongylid egg counts were conducted for each animal and egg per gram counts were used to rank individuals over the winter housing and grazing season. The mean rank was used to select animals for future breeding use. Sires were initially selected from a pool of 83 available males on the basis of low FEC over one grazing season prior to this study (Fig. 1). During the study sires were chosen using the same method from the selected yearling group with three sires being used annually. Older or ill females from the breeding herd were replaced annually with female yearlings, again based on ranked low egg shedding. Three subgroups were created within the breeding herd and segregated during mating so that inbreeding was avoided, with one sire being used for each group. The same number of kids as in the selected group was chosen at random from the wider herd and formed the control group. These were co-grazed and housed with the selected line throughout the study period.

2.3. Study design

Approximately 80 male and 80 female experimental animals from either single, twin or triplet birthings, split equally between selected and controls, were monitored on a five weekly basis from the end of the first season at approximately 5–6 months old, through winter housing and turnout onto pasture in May and through the summer grazing season until the animals were housed in late September. Animals were infected naturally through pasture grazing, but also received a single dose of 10,000 drug susceptible *T. circumcincta* L_3 towards the end of the winter housing period. For the analysis, data was analysed within four defined time windows; (1) the end of the first grazing season, when the animals had some exposure to parasite infection, but were still with their dams and thus had milk available, (2) early in the second season prior to turnout during the period when animals were artificially infected with a single dose of infective nematode larvae, (3) early season grazing post turnout and pre-weaning and (4) from weaning until winter housing.

2.4. Parasitological sampling and analysis

Faeces were sampled directly from the rectum at each of the five weekly sampling point and faecal egg counts (FEC) performed using a saturated salt flotation method as described by Christie and Jackson (1982) with a sensitivity of 1 egg per gram (epg).

2.5. Anthelmintic treatment and performance analysis

Anthelmintic treatment was administered to all animals at each sampling point, either fenbendazole or ivermectin commercial formulations at the manufacturers recommended dose rate in line with farm practice. Samplings were carried out on a five weekly rotation designed to ensure complete clearance of the previous treatment and allow sufficient time for new infections to become patent. In addition to birthweights being recorded, animals were also weighed at this time using a spring balanced weigh-crate as part of the routine performance monitoring procedures carried out by the farm. F9 male data is restricted to third and fourth window as the animals were not weighed during the first and second window.

2.6. Blood and plasma sampling

Blood samples were collected from the jugular vein into a Vacutainer tube® (Beckton Dickinson) containing anticoagulant (heparin or sodium EDTA) at each sampling point. Farm access restrictions during the F7 generation due to a UK Foot and Mouth disease epidemic prevented blood sampling, so only one sample was available for this generation. Aliquots of whole blood were removed for eosinophil counts prior to centrifugation at $1750 \times g$ for 15 min, following which plasma was removed and collected into sample cups for storage at $-20\,^{\circ}C$ until required for analysis.

2.7. Eosinophil counts

Fifty microlitre aliquots of whole blood were added to 450 μl of freshly prepared Carpentier's stain and mixed using a vortex mixer. Cells were counted under a compound microscope at ×100 using Fast-Read 100 slides (Immune Systems Ltd.) and the results expressed as cells per μl.