Comparative survival of first-stage larvae of small lungworms *Muellerius capillaris* and *Neostrongylus linearis* of goats in alfalfa and ryegrass plots

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Summary — Faeces from a goat infected with *M* capillaris and from another infected with *N* linearis were used to assess survival of first-stage larvae under natural conditions in northwest France in mid-autumn, at the high-risk period. The faeces were deposited on 3 plots covered respectively with ryegrass, short cut alfalfa and high alfalfa. The desiccation of faeces and survival of larvae were tested at 1, 7, 14 and 21-d intervals. Survival of *M* capillaris was much higher than that of *N* linearis in all plots. The lower desiccations were observed in the high alfalfa plot whereas the best survivals were found in the 2 other plots. Vegetation where faeces are deposited may thus play a part in the accumulation of parasitic risk on a pasture.

*R* Muellerius / *Neostrongylus* / vegetation / first-stage larvae / survival

Résumé — Comparaison de la survie des larves du premier stade des protostrongyles *Muellerius capillaris* et *Neostrongylus linearis* d’origine caprine sur des parcelles de luzerne et de ray-grass. Les fèces de 2 chèvres, l’une infectée par *M* capillaris et l’autre par *N* linearis, ont été utilisés pour estimer la survie des larves du premier stade dans les conditions naturelles du nord-ouest. L’essai a eu lieu à la mi-automne, qui constitue la période à haut risque. Les fèces ont été déposées sur 3 parcelles respectivement couvertes par du ray-grass, de la luzerne coupée courte ou haute. Le dessèchement des fèces et la survie des larves ont été mesurés 1, 7, 14 et 21 j après les dépôts. La survie de *M* capillaris a été très supérieure à celle de *N* linearis sur les 3 parcelles. Un dessèchement moins important a été enregistré sur la parcelle avec de la luzerne haute, alors que les survies des larves étaient supérieures sur les 2 autres parcelles. La végétation des parcelles sur lesquelles les fèces sont déposées peut jouer un rôle dans l’accumulation du risque parasitaire sur le pâturage.

*R* Muellerius / *Neostrongylus* / végétation / larve du premier stade / survie

* Correspondence and reprints
INTRODUCTION

Protostrongylid nematodes (small lungworms) have a 2-host life-cycle in which small ruminants serve as definitive host and molluscs as intermediate hosts. The first-stage larvae (L₁) are the only non-parasitic stage and their resistance to climatic harshness is thus important for the completion of the life-cycle. The survival of L₁ of *M. capillaris* in sheep faeces outdoors has been studied in England (Rose, 1957), Spain (Requena-Feo, 1983) and Morocco (Cabaret, 1986). The latter 2 authors found that survival of L₁ was at its highest in October and November. No data are available for L₁ of *N. linearis* outdoors. Survival of L₁ of protostrongylids of goat origin has never been assessed and may differ from those of sheep origin.

The height and type of vegetation in natural pastures of north-western France (Touraine) is an important factor regulating desiccation of deposited sheep faeces (Mauldon and Gruner, 1984). It has been demonstrated that desiccation of sheep and goat faeces might be the key-factor to L₁ survival in experimental conditions (Requena-Feo et al, 1986; Cabaret et al, 1991). This fact could in part explain why a humid period such as autumn is the high-risk period for goat infection in Touraine (Cabaret, 1983; Richard et al, 1990). Artificial pastures are common for goats in this area and are often sown with alfalfa (*Medicago sativa* L) or ryegrass (*Lolium perenne* L). The purpose of this paper is to estimate the survival of L₁ of *M. capillaris* and *N. linearis* in relation with goat faeces desiccation in alfalfa and ryegrass plots at a high-risk period for goat infection, ie autumn.

MATERIALS AND METHODS

The faeces were collected from 1 goat naturally infected with *M. capillaris* and another artificially infected with *N. linearis*. The average excretion was 1 229 ± 167 L₁/g of faeces (LPG) for the former and 37 ± 10 for the latter. They were kept indoors and fed on a diet of hay and commercial pellets. The plots of alfalfa (2 x 1200 m²) and ryegrass (1 200 m²) were included within a larger pasture of ryegrass (13 300 m²) located at the INRA research center in Nougilly (France). The pasture was normally cut for hay production. The plots were cut at the end of July and in September. The height of grass was adjusted to 15-20 cm for ryegrass and short alfalfa and to 45-50 cm for high alfalfa at the start of the experiment.

The faeces were collected every morning and deposited on each plot by group of 3 replicates for each species before 11 am. The replicates weighed respectively 5 and 7 g for *M. capillaris* and *N. linearis*. Each replicate was put in a grey plastic cylindrical device 3 cm high and 6 cm in diameter with a white plastic grid at the bottom (mesh: 5 μm) that allowed contact with grass but hindered migration of L₁ from faeces onto the plot. Another group of 3 replicates was used to assess the number of L₁ and the dry matter (DM) of freshly collected faeces and was used as controls. The faecal deposits remained during various periods of time on the plots: 1) 1 d (repeated every d from 1-7 November 1989); 2) 7 d (21-28 November); 3) 14 d (17 November-2 December); 4) and 21 d (11 November-2 December).

Dry matter of faeces was estimated on 10 g of faeces maintained at 100 °C for 24 h (1-[weight of fresh faeces — weight of desiccated faeces]/weight of fresh faeces). Survival was calculated as number of larvae in fresh faeces — number of larvae in faeces deposited on plots/number of larvae in fresh faeces. The meteorological data were obtained from a station located at 50 m from the plots.

The analysis of data was performed with the Stat–Itc statistical computer package (1988) for analysis of variance (ANOVA and Newman–
Keuls test) and multiple analysis of variance (MANOVA; significance with Wilk’s test). A lethal time when 50% of L₁ population died was calculated using a log-probit computer software (Raymond, 1985).

RESULTS

Climatological data and desiccation of faeces

The averages, minima and maxima of mean daily temperatures for the various periods of investigations are shown in figure 1. The temperatures were very similar for 7-, 14- and 21-d trials (range of averages of mean temperatures: 4.2–6.7 °C) but were hotter for 1-d trials (10.8 °C). Rainfall was 1.6, 1.8 and 4.1 mm respectively during 7-, 14- and 21-d trials. Dry matter of faeces deposited in plots versus control is shown in figure 2. The confidence intervals ranged from 1.1–7.9% at P = 0.05. The controls were never significantly different from one period to another. On daily deposits, the faeces from the goat infected with M capillaris were less desiccated than those of the N linearis-infected goat (fig 2). The dry matter of faeces from both goats did not differ significantly later on in 7- to 21-d trials. The faeces located in high-alfalfa plots were less desiccated than those located in short-alfalfa or rye-grass from 7 to 21 d after deposition.

Survival of L₁ and vegetation (fig 3)

Survivals appear to be similar for the 3 types of vegetation but slightly lower survivals are recorded in high-alfalfa plots for both species. From the ANOVA on factor vegetation it was concluded that: 1) no significant difference was established on d 1; 2) on d 7, 14 and 21 post-deposition, L₁ in faeces on ryegrass and short-alfalfa plots had a significantly better survival than on the high-alfalfa plot. The lethal times when 50% of larvae died (LT₅₀) are shown in table I. The LT₅₀ was at its highest for M capillaris in ryegrass.

![Graph showing daily temperature °C](image-url)

**Fig 1.** Minimum, maximum and average daily temperatures during the 1-, 7-, 14- and 21-d experiments.
M. capillaris

N. linearis

Fig 2. Evolution of faecal dry-matter in relation to vegetation and time: M capillaris and N linearis.
A multivariate approach to L₁ survival

L₁ survivals were dependent upon species of protostrongylid and vegetation at the various sampling dates and from MANOVA the following was obtained: the sampling date was the most important factor of variation ($F (3.10) = 114.6; P > 0.999$); this factor interacted with vegetation ($F (6.20) = 3.5; P > 0.98$) and with protostrongylid species ($F (3.10) = 10.0; P > 0.99$); this means that evolution in time depended on vegetation and species of protostrongylid; the interaction species x vegetation was dependent on sampling dates ($F (6.20) = 2.6; P > 0.95$); it shows that species did not behave the same way in relation with vegetation at the various sampling dates.

**Table I.** Times at which 50% of *M. capillaris* (M) and *N. linearis* (N) first-stage larvae died (LT₅₀) in relation with vegetation.

<table>
<thead>
<tr>
<th>Vegetation of plots</th>
<th>Protostrongylid species</th>
<th>LT₅₀ (d)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ryegrass</td>
<td>M</td>
<td>27.0ᵃ (12.1–62.0)ᵇ</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>2.4     (1.2–3.7)</td>
</tr>
<tr>
<td>Alfalfa (short)</td>
<td>M</td>
<td>13.7    (6.4–22.2)</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>1.0     (0.2–2.1)</td>
</tr>
<tr>
<td>Alfalfa (high)</td>
<td>M</td>
<td>10.0    (7.1–14.0)</td>
</tr>
<tr>
<td></td>
<td>N</td>
<td>2.3     (1.4–3.3)</td>
</tr>
</tbody>
</table>

ᵃ Mean lethal time;ᵇ confidence interval ($P > 0.95$).

**DISCUSSION**

The ability of *M. capillaris* L₁ to survive in goat faeces is comparable to other records in sheep. The survivals at 1, 2 and 3 wk post-deposition were slightly lower than those recorded by Rose (1957) in December in England and slightly higher than those found by Reguera-Feo (1983) in November and December in Spain or by Cabaret (1986) in November in Morocco. *N. linearis* had lower survival rates than *M.

Fig 3. Comparative survivals of *M. capillaris* and *N. linearis* in ryegrass, short and high alfalfa plots.
capillaris, as previously demonstrated in experimental conditions (Cabaret et al., 1991; Morrondo-Pelayo et al., 1992).

The vegetation in which faeces are deposited influenced their desiccation, as already shown in the same region (Mauléon and Gruner, 1984). The faeces in high-alfalfa plot remained more humid than in other plots. In our conditions, desiccation of faeces was not the key-factor to survival: ryegrass and short-alfalfa plots allowed high survivals although desiccation was significant (up to 72% DM). This is in apparent contradiction with experimental findings (Morrondo-Pelayo et al., 1992) where DM of faeces were related to survivals of L₁. The experimental desiccations were performed rapidly (in less than 24 h) and were more extreme (up to 95%) and thus resembled more closely climatic conditions prevailing in summer than in autumn. The indirect effect of vegetation on L₁ survivals might be searched in the interaction between desiccation and temperature as has been previously shown in experimental conditions (Cabaret et al., 1991). The absence of N. linearis in natural infection of goats (Cabaret, 1983) is probably due in part to low survival in faeces at the L₁ stage.

In the temperate climate of northwestern France, autumn is the period at high-risk for protostrongylid infection (Cabaret, 1983): goats excrete more larvae in faeces and land-snails and slugs are still in activity. The good survival of M. capillaris L₁ at this season is an additional high-risk factor (for molluscs and subsequently goats) and the use of different types of pastures (ryegrass and short alfalfa vs high-alfalfa) may modify only slightly the risk.

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REFERENCES


Reguera-Feo A (1983) Sobre la epizootiologia de las protostrongiloidosis ovinas en la provincia de León. Tesis Doctoral Fac. Biología, Univ. León (Spain), 1-200


