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## SEX- AND AGE-SPECIFIC LUNGWORM INFECTION IN ROCKY MOUNTAIN BIGHORN SHEEP DURING WINTER

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**ABSTRACT:** Sex- and age-specific fecal lungworm (*Protostrongylus* spp.) larvae concentrations in Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*) from the Cinnabar Winter Range in southwestern Montana were determined. All 228 fecal samples collected from bighorn sheep of known sex and age class from November to April, 1984 to 1987 contained first-stage lungworm larvae. Fecal lungworm concentrations of ewes and rams declined significantly from late fall through early spring, whereas number of lungworms in lamb feces increased as winter progressed.

**Key words:** Larvae, lungworms, *Protostrongylus* spp., Rocky Mountain bighorn sheep, *Ovis canadensis canadensis*, stress.

### INTRODUCTION

Lungworms (*Protostrongylus* spp.) have been implicated as proximate, ultimate, and complementary factors in many epizootics of Rocky Mountain bighorn sheep (*Ovis canadensis canadensis*) (Buechner, 1960; Forrester and Senger, 1964; Wishart et al., 1980). Spraker (1979) found substantial lamb mortality associated with lungworm infection in a Colorado bighorn herd. Lungworms may influence health and fitness of adult bighorns, but signs may be less acute than in lambs.

Differences in intensity of lungworm parasitism among sex and age classes of bighorn sheep have been reported by Blood (1963), Forrester and Senger (1964), Thorne et al. (1976), and Festa-Bianchet (1987). However, statistical differences associated with seasonal and sex and age interactions of lungworm infection usually have not been addressed. Our objective was to compare the fecal lungworm concentrations during three winters for lambs, ewes, and rams from a bighorn population wintering in southwestern Montana.

### MATERIALS AND METHODS

The Cinnabar Winter Range (CWR) (45°04'N, 110°44'W) is located 10 km north of Gardiner, Montana (USA). The CWR covers approximately 350 ha on the west side of the Yellowstone River (Keating et al., 1985) with elevations

ranging from 1,500 to 1,900 m. Annual temperature and precipitation on the study area from 1951-71 averaged 5 C and 27.5 cm, respectively (Keating, 1982). The CWR has an east to southeast aspect and remains relatively snow-free during most winters.

Between 80 and 150 bighorn sheep typically occupy the CWR from early November to early April. These sheep are habituated to humans and often allow approach to within 5 m. The CWR is accessible to the public, and human activity (vehicles, photographers, research, ranching, hunting, and domestic livestock grazing) is common throughout the period when sheep occupy the area.

The study was conducted between early November and early April, when sheep occupied the winter range, from 1984 to 1987. Fresh fecal samples were collected from the ground after observing sheep of known sex and age defecate. Sheep were classified either as lambs (<1.5-yr-old), adult ewes (≥1.5-yr-old), or rams (≥1.5-yr-old). Fecal pellets were stored in plastic bags at 9 C until analyses. All samples were analyzed within 12 days of collection.

A modified version of the Baermann technique (Baermann, 1917) was employed to extract first-stage lungworm larvae from feces. Pellets were weighed to the nearest 0.1 g, slightly crushed, and soaked in water in a Baermann apparatus for 24 hr (Beanne and Hobbs, 1983). Larvae were counted under a dissecting microscope and expressed as first-stage larvae per gram of feces (LPG).

We used simple and polynomial regressions to examine the relationships between LPG and date of fecal collection for lambs, ewes, and rams. Outliers were identified with Cook's distance measure (Neter et al., 1989) and excluded

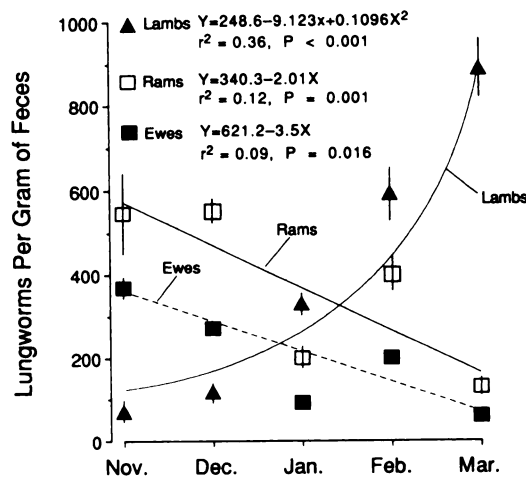


FIGURE 1. Concentration of lungworms in feces of lambs, ewes, and rams from early November to early April, 1984–87, at the Cinnabar Winter Range, Montana. Vertical lines through monthly means are  $\pm 0.25$  SE of the mean.

from analyses. Significant second-order coefficients in polynomial regression analyses were used to identify non-linear relationships (Neter and Wasserman, 1974). Based on residuals from these regressions, we noted strongly heteroscedastic variances, typical of lungworm data (Uhazy et al., 1973), for each sex and age group; thus, all subsequent analyses were conducted on  $\log_{10}$  transformed lungworm concentration estimates. Logarithmic transformations also convert nonlinear data into a linear form (Zar, 1974), further improving suitability for linear parametric statistical analyses.

We tested for differences in slopes and y-in-

tercepts of regression equations for lambs, ewes, and rams using the General Linear Models procedure of the Statistical Analysis System (SAS Institute Inc., 1988), with data pooled among years. Sample sizes within years were too small for meaningful comparisons among years; however, we investigated the consistency of lungworm-season trends among years using simple regression within sex and age classes.

## RESULTS

We analyzed 228 fecal samples collected between November and April, from 1984 to 1987. All fecal samples contained first-stage larvae of the genus *Protostrongylus*. Mean ( $\pm$ SE) LPG for lamb, ewe, and ram feces were 234 ( $\pm 51$ ), 112 ( $\pm 17$ ), and 194 ( $\pm 34$ ), respectively. Mean fecal lungworm concentrations in lamb feces increased from 70 to 890 LPG from early November through early April (Fig. 1). In contrast, mean LPG in ram feces declined from 545 to 130, and ewe LPG declined from 370 to 58. Based on polynomial regression analyses, the significant increase in LPG of lambs was nonlinear ( $P \leq 0.001$ ,  $r^2 = 0.36$ ), whereas the significant declines in LPG were linear ( $P = 0.016$ ,  $r^2 \geq 0.09$ ) for rams and ewes (Fig. 1).

The LPG trends observed (Fig. 1) generally were consistent among years for each sex and age class (Table 1). Regression equations were similar among years, and almost all were either significant ( $P \leq 0.05$ )

TABLE 1. Simple regression statistics of lungworm larvae per gram of feces (LPG), transformed using  $\log_{10}$ , and day of sampling by year for rams, ewes, and lambs.

Sex/age	Year	Equation*	SE	n	$r^2$	P
Ad. rams	1985	LPG = 7.8 - 0.034 (day)	0.012	12	0.43	0.020
Ad. rams	1986	LPG = 4.0 + 0.013 (day)	0.007	18	0.15	0.116
Ad. rams	1987	LPG = 6.6 - 0.020 (day)	0.006	35	0.27	0.001
Combined		LPG = 5.7 - 0.010 (day)	0.005	65	0.07	0.037
Ad. ewes	1985	LPG = 5.5 - 0.015 (day)	0.008	27	0.11	0.090
Ad. ewes	1986	LPG = 5.0 - 0.011 (day)	0.004	50	0.15	0.006
Ad. ewes	1987	LPG = 6.2 - 0.023 (day)	0.003	41	0.60	0.001
Combined		LPG = 5.4 - 0.016 (day)	0.003	118	0.25	0.001
Lambs	1985	LPG = 3.7 + 0.022 (day)	0.010	7	0.46	0.092
Lambs	1986	LPG = 3.4 + 0.029 (day)	0.006	15	0.60	0.001
Lambs	1987	LPG = 3.8 + 0.017 (day)	0.004	23	0.51	0.001
Combined		LPG = 3.6 + 0.022 (day)	0.003	45	0.53	0.001

\* Day = collection date.

or nearly significant ( $P < 0.10$ ) each year for both ewes and lambs. The regression equation for rams in 1986, however, differed substantially from equations in 1985 and 1987 and was not significant ( $P = 0.116$ ).

There were significant differences in the slopes of regression curves between ewes and lambs ( $P \leq 0.001$ ) and between rams and lambs ( $P \leq 0.001$ ). Slopes did not differ significantly between ram and ewe regression curves ( $P = 0.230$ ). But there was a significant difference in mean LPG between ewes and rams ( $P = 0.002$ ).

### DISCUSSION

Differences in fecal lungworm concentrations between lambs and adults generally were consistent during all three winters of the study. The substantial increases in LPG of lambs, relative to adults, from fall to spring may have been due to higher susceptibility of lambs to winter weather and poor forage conditions. Immunocompetence is contingent upon nutritional status, particularly in juveniles (Chandra, 1988). Also, infection by lungworms after consumption of infected snails in fall/winter may increase the passage of larvae in lambs (Lange, 1973; Samson et al., 1987) but not in adults (Hibler et al., 1982; Festa-Bianchet, 1987).

Fecal lungworm concentrations of adult rams and ewes declined through winter, but LPG in ram feces averaged nearly twice that of ewes. Increased LPG in rams may have been due to higher levels of stress experienced during the rut (Geist, 1971; Festa-Bianchet, 1987), perhaps exacerbated by extensive human activity and weather conditions on the CWR at this time. Competition among rams also may have reduced immunocompetence due to stress-induced elevated serum cortisol levels (Harlow et al., 1987).

The linear decline in fecal lungworm concentration of adults from fall to spring differed considerably from other studies reporting peak LPG in winter or spring

(Forrester and Senger, 1964; Uhazy et al., 1973; Festa-Bianchet, 1987). We are unsure why this pattern occurred, but substantial declines in human activity, reduced group sizes, and moderating weather conditions after early winter may have resulted in less stress on adult sheep. Increased stress has been associated with higher fecal lungworm concentrations in bighorns (Fougieri-Tower and Onderka, 1988). Perhaps decreased stress contributed to the decrease in larval shedding we observed in adult sheep.

Lungworms, and their role in bighorn sheep population dynamics, continue to present managers and researchers with many perplexing questions. Failure to identify sex- and age-specific differences in fecal lungworm concentration could bias the interpretation of lungworm infection data. Variation in lungworm concentration among sex and age classes of bighorn sheep may reflect differences in susceptibility to parasitism or individual health (Festa-Bianchet, 1987). Future observations of lungworm parasitism should address sex- and age-specific variation among bighorns to verify our findings. We believe various types of stress may deteriorate the ability of bighorns to suppress lungworm parasitism, thus affecting individual productivity. Studies are needed to identify the role of stress in lungworm infection and how it may influence individual bighorns of different sex and age. Questions about stress-induced immunosuppression and its effects on larval output, longevity of adult lungworms, and intensity of infection in the lungs relative to fecal lungworm concentration should also be addressed by researchers.

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