13 April 1977

Kate Wynne 248 B North Ashury St. Moscow, Idaho 83843

Dear Kate:

It's a pleasure to inform you that your proposal entitled "An Investigation of the Intermediate Hosts of Protostrongyline Nematodes on Two Bighorn Winger Ranges in Idaho" has been accepted by the Wilderness Research Center. We hope you can begin on this project at least by early June. We will expect you to register for 3 credits of special problems, FWR 499, for the summer. The project report will be due in polished draft form five weeks after the start of the fall semester. We then expect the reports will be ready for publication by the end of fall semester.

Your \$600 honorarium will be paid upon completion of the summer field work and in time for fall registration. Arrangements for food, lodging or transportation whould be made through your project advisor and Mr. Ken Sowles.

Congratulations on a well-written proposal.

Sincerely,

John H. Ehrenreich Dean

JHE: bmk

An Investigation of the Intermediate Hosts of Protostrongyline Nematodes on Two Bighorn Winter Ranges in Idaho

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Kate Wynne March 25, 1977

# INTRODUCTION

Bighorn sheep are "seriously controlled by disease". This is the conclusion reached by Forrester and Senger (1963) and others who have studied wild bighorn populations. Among the numerous bighorn diseases, pneumonia has been recognized as a frequent and significant vector of mortality. The effects of this disease are accentuated when the infected populations are stressed by dietary or mineral deficiancies (Blood, 1963 and Fackard, 1946). The contraction of the pneumonia-causing bacteria <u>Pasteurella ovisepta</u> is often facilitated by irritation and congestion created by several lungworm species (Protostrongyline nematodes)that imbed themselves in the parenchymal and bronchiolar lung tissues (Packard, 1946).

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It is believed that the existence of these parasitic nematodes is dependent on the presence of a sheep host (<u>Ovis canadensis</u>) and an intermediate host, thought to be terrestrial snails (Marsh, 1965). Although lungworm breeding and early development occur in the sheep host, development into an infective larva must occur in the intermediate host. Thus, the ecology and life history of the intermediate host snails become a focal point in the study of the proximate factors of mortality among bighorn sheep.

Numerous lungworm-related studies have been carried out in Montana, but little data has been obtained from Idaho ranges concerning the source and extent of lungworm infestation in Idaho bighorn herds. Couey (1950) conducted a study on the fecal evidence of lungworm larvae in Montana bighorn herds and reported 75-100% infestation. Pillmore (1958) observed that lungworm infestation on Colorado ranges was directly related to the presence of terrestrial snails which appear to act as intermediate hosts for these nematodes. Senger and Forrester (1962) identified several snails as possible intermediate hosts for <u>Protostrongylus stilesi</u> on Montana ranges but similar data for Idaho ranges is limited or nonexistent. Smith (1954) collected lungworm data from 229 fecal specimens from Salmon River bighorn ranges and found that 50% of the specimens contained lungworm larvae. The species of snails that facilitate the infestation of sheep by these nematodes are largely unknown.

In brief, lungworm infestation has been determined to be a significant proximate cause of death among bighorn populations and is prevalent in the Idaho herds that have been studied. This infestation is believed to be dependent on intermediate host snails which are ingested by bighorn sheep. To date, in Idaho, these intermediate host species have not been identified and can only be assumed to be similar to those which occur on Montana ranges. The lungworm host species present on Idaho ranges must be identified before lungworm infestation, and subsequent bighorn mortality, can be understood and managed on these ranges.

The objectives of this study are to: 1) determine the incidence and frequency of lungworm larvae in various terrestrial snails on two winter ranges, 2)determine the density and frequency of host snails on these ranges, and 3)discern the extent to which sheep of these ranges in the Idaho Primitive Area are infested by lungworms.

# STUDY AREA

This study will be based at the Taylor Ranch of the University of Idaho's Wilderness Research Center on Big Creek in the Idaho Primitive Area of central Idaho.

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Samples will be collected during the summer of 1977 on two bighorn sheep wintering grounds. One range is located directly across from Taylor Ranch on Big Creek and another is located on the east side of the Middle Fork of the Salmon River, just west of the Waterfall Creek trail.

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#### METHODS

Land snails and pellet groups will be collected and analyzed for each of two winter ranges. Samples will be collected on these ranges and analyzed at the Taylor Ranch facility. Results obtained from the two ranges will be compared to determine whether significant differences occur between infestation levels and host species on these ranges.

Snail Collection

Snails will be collected on both ranges early in the study, prior to pellet collection and sample analysis. By performing this sampling prior to the onset of the hot and dry conditions of midsummer, sampling bias will be limited by insuring snail activity in all vegetation types.

Terrestrial snails will be collected on the two designated ranges at 1000 foot intervals between 4000' and 6-or 7000' elevations. Transect lines will be plotted approximately 30 meters apart and will intersect all possible cover types. Ten transect lines onleach range ten will be randomly selected and  $_{\Lambda}$  one-m<sup>2</sup>-quadrats will be placed along them in well defined cover types at ten-pace intervals. Where applicable, the duff and humus layers within these plots will be filtered through a 1 or 2 mm mesh screen to expose snails which dwell under needles, leaves, and decaying material. Live specimens will be placed in vials and classified according to the elevation, vegetation type, and microenvironments they were found in. Empty shells will be similarly classified but will be packed in cotton prior to transport in vials. All specimens will be taken to the Taylor Ranch facility for analysis.

The snails and shells collected will be identified according to Morton (1967) and Burch (1962) and the frequency and relative density of snails present in various cover types will be determined by averaging the number found per square meter. Live snails will be examined for the presence of lungworm larvae by the technique described by Forrester (1962). A snail is placed on the inside of a petri dish and sprayed with a fine water mist so it will crawl across the glass surface. As it begins crawling, the petri dish is inverted and placed under a dissecting scope. Dark "half-moon shaped objects" within the snail's foot indicate the presence of larvae. These larvae will be counted, extracted, and identified according to Yorke and Maplestone (1962). The snails that contain larvae will be recorded by species, number of larvae contained, and site conditions from which they were collected. The frequency of lungworm infestation in each snail will be recorded and the relative density of infected snails on the ranges will be determined by averaging the number of infested snails found per square meter.

## Pellet Group Collection

Pellet groups will be collected to determine the per cent incidence of infestation in the sheep present on two winter ranges. Fresh pellet groups will be collected soon after defecation is observed to eliminate pellet identification problems. The groups collected will be catagorized according to the vegetation type and

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altitude they were deposited at, and the sex and age group of the sheep that deposited them.

Pellet groups will be taken to the Taylor Ranch facility for analysis and determination of the incidence of lungworm infestation. When dry, samples will be weighed and run through a Baermann apparatus (Fig.1) to determine which pellet groups contain larvae, and at what density (larvae/gram dry feces). The percent of samples from each range showing evidence of lungworm infestation will be recorded. Identification of nematode larvae will be based on Yorke and Maplestone (1962).

#### SUMMARY

This project involves field collection and laboratory analysis of fecal pellets and terrestrial snails. Fecal sample analysis will facilitate estimation of lungworm infestation in the sheep present on two winter ranges. The degree of infestation within these individuals will be estimated (relative density of lungworm larvae per sample) and the protostrongyline species present will be determined.

Collection of terrestrial snails will be followed by a search for evidence of lungworm infestation. Data collected from this search will be utilized to decide which species of snails are the most frequent protostrongyline hosts on these two ranges. Finally, estimates of the relative density and location of these snail hosts will be determined.

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Fig.1 BAERMANN APPARATUS: Fecal samples are wrapped in cheesecloth and placed on a screen in a water-filled glass funnel for 24 hrs. Nematode larvae present in the feces are activated by heat and moisture, emerge from the feces, and sink to the stopcock at the bottom of the funnel. After 24 hrs, the larvae are released into the beaker where they may be removed with a pipette for analysis.

#### Literature Cited

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