

Domesticated Birds Take Down *P. tenuis*

Morgan Linn

Introduction

Parelaphostrongylus tenuis (*P. tenuis*) is a common nematode of white-tailed deer (*Odocoileus virginianus*) that requires ingestion of an infected intermediate host (gastropod), for transmission to its definitive host (McCoy and Nudds, 2000). White tailed deer infected with *P. tenuis* exhibit no clinical signs (Samuel et al., 1992), but *P. tenuis* can become deadly for other ungulates (Pennsylvania Game Commission), in this case goats.

Goat grazing has become an increasingly popular way to combat invasive species such as buckthorn. Although goats are a great way to combat invasive plant species, in areas that contain white tailed deer, *P. tenuis* poses a threat to these goats often resulting in death.

Many suggest birds for pest reduction. One proposed hypothesis by Brooks (2016) to prevent infections in goat herds, is to introduce hens to decrease gastropod abundance. Many suggest birds for pest reduction, but little is known about the effectiveness of poultry on gastropods. The perception of biological motion is congenital of the vertebrate visual system (Johansson, 1973), more specifically newborn chicks show an innate preference for biological motion displays (Vallortigara et al. 2005; Vallortigara and Regolin, 2006). This is relevant because moving objects may represent insects which are the natural prey of many species of poultry. Allowing poultry to graze before and during goat grazing may reduce the abundance and diversity of gastropods present, therefore reducing the likelihood that a goat may become infected. However, the efficiency is unknown.

I will be analyzing the gastropod abundance and diversity before and after poultry and goat grazing. I hypothesize quadrats that have received a ‘treatment’ of poultry prior to goat

grazing will have significantly reduced abundance and diversity of gastropods, compared to fields that only experienced goat grazing.

Methods

The study area was in Rosemount, MN. There was a control area (no poultry or goats), an area that was solely grazed by goats, and an area that was first grazed by poultry and then grazed by poultry and goats. There were three transects in each study area, from which leaf litter and the top 2cm of soil was collected from a standardized quadrat along the transects. Once samples were collected, the leaf litter and top soil was sorted through a series of sieves, to search for gastropods. The gastropods were placed in the refrigerator until further examination. A dichotomous key to Wisconsin terrestrial snails created by Dr. Kathryn E. Perez, was used to identify the species.

After identification, the gastropods were artificially digested in a solution of hydrochloric acid (HCL), water, and powdered pepsin. Tap water (166ml) was placed in a large beaker, which was then placed in a fume hood where 1.3ml of HCL was added to the water while stirring. Lastly, 1 gram of powdered pepsin was added to the solution. The gastropods were then grouped by species and placed in a flat-bottomed container containing the solution, such as a petri dish, for about 16 hours at 35-40 degrees Celsius to simulate mammal digestion. After digestion, the petri dishes containing the gastropods were examined for nematodes with a dissection microscope.

All data analysis was completed in R studio (version 1.1.456). Information from Lankester (2001) regarding whether species are known intermediate hosts was added to the data set. The quadrat data was then combined to create transect data and raw values were converted to

density. The data was then stratified first by native or exotic species and then by intermediate host status (yes or no).

To calculate richness and diversity indices, individuals of each species per transect were added up and put into a site by species matrix. Shannon's diversity index, Simpson's diversity index, and species richness were all calculated using the vegan package (Oksanen et al. 2019). The treatment type and timing (before or after treatment) were added to each index.

After that, a non-metric multidimensional scaling (NDMS) ordination was done using the vegan package (Oksanen et al. 2019) and code from Grieger 2019, to identify patterns in the data. Multivariate GLM's were then used to examine Shannon's and Simpson's diversity indices, as well as richness with all factors. A Poisson regression was used because we had count data. `mvabund` was then used for multivariate generalized linear model's (GLM). Lastly, an ANOVA was then run using negative binomial to determine the effects between species.

Results

Figure 1 is graphing gastropod count data; it suggests that after treatment, all treatments have an increase in gastropods ($p = 0.674$ after bird treatment and 0.006 after goat treatment). The goat treatment has the highest difference in gastropod counts between treatments. Figure 2 is showing specifically exotic slugs to Minnesota using density and it shows that treatments have the opposite effect. It suggests that density decreases after treatment. Figure 3 shows intermediate hosts of the parasite using density ($p = .104$ after bird treatment and 0.072 after goat treatment). Density of intermediate hosts appears to increase after treatment of goats, but decrease after birds, but this was not statistically significant.

The Shannon diversity index shows that diversity increases slightly after treatments ($p = 0.476$ after birds and 0.516 after goats). The Simpson inverse index had a similar trend ($p =$

0.651 after birds and 0.490 after goats), as well as species richness ($p = 0.399$ after birds and 0.353 after goats).

The NMDS stress value was 0.1508, meaning it is a good representation of goodness of fit in the reduced dimension. Figure 7 displays hulls enclosing points of each treatment type that show how the treatments differ due to timing. Figure 9 shows the species that have the largest effect on the site distribution pattern, those species are *Vitrina limpida*, *Cochlicopa lubricella*, *Deroceras reticulatum*, *Nesovitrea electrina*, *Vallonia costata*, and *Zonitoides arboreus*. *V. costata* and *N. electrina* appear to increase after both treatments.

Figure 14 shows the GLM after only considering intermediate hosts. Figure 12 shows abundance using the R package mvabund (Wang et al. 2020).

We did find several nematodes after dissections, pictured in the appendix. The nematode found in a *Deroceras reticulatum* is possibly *P. tenuis*.

Discussion

We are mostly interested in the poultry grazing effect on intermediate hosts because they are needed to complete the life cycle of *P. tenuis*. An interesting observation is that the goat treatment has the largest increase in number of gastropods, whereas birds seem to have no effect; however, the density of exotic and intermediate hosts appears to decrease substantially after the bird treatment.

Although there is a clear trend towards the reduction of intermediate hosts after the grazing of poultry, we can see that by the size of many error bars, how hulls in figure 8 overlap, and the size of our p-values from the GLM's, that our results are not statistically significant. This may be due to our small sample size. However, our results are very interesting and call for a

larger study in the future. A future study should include multiple study areas with more transects and treatments.

References

- “Brain Worm.” *Pennsylvania Game Commission*,
www.pgc.pa.gov/Wildlife/WildlifeRelatedDiseases/Pages/BrainWorm.aspx.
- Brooks, Kelly S. “Meningeal Worm in Central Iowa Goat Herds.” *Iowa State University*, vol. 662, no. 1, 2016.
- Grieger, R. 2019. NMDS Ordination. RPubS. <<https://www.rpubs.com/RGrieger/545184>> Accessed Apr 2020.
- Lankester, M. W. 2001. Extrapulmonary Lungworms of Cervids. *Parasitic Diseases of Wild Mammals* 228–278.
- McCoy, Karen D, and Thomas D Nudds. “An Examination of the Manipulation Hypothesis to Explain Prevalence of *Parelaphostrongylus Tenuis* in Gastropod Intermediate Host Populations.” 2000.
- Oksanen, J., F. Blanchet, M. Friendly, R. Kindt, P. Legendre, D. McGlinn, P. Minchin, R. O'Hara, G. Simpson, P. Solymos, M. Henry, H. Stevens, E. Szoecs, and H. Wagner. 2019. <https://CRAN.R-project.org/package=vegan>
- Perez, K. E. n.d. Dr. Kathryn E. Perez. <<http://northamericanlandsnails.org/snailresources.html>> Accessed Jan 2020.
- Vallortigara, Giorgio, and Lucia Regolin. “Gravity Bias in the Interpretation of Biological Motion by Inexperienced Chicks.” *Current Biology*, vol. 16, no. 8, 18 Apr. 2006.
- Vallortigara, Giorgio, et al. “Visually Inexperienced Chicks Exhibit Spontaneous Preference for Biological Motion Patterns.” *PLOS Biology*, 7 June 2005.
- Wang, Y., U. Naumann, D. Eddelbuettel, J. Wilshire, and D. Warton. 2020. <<https://CRAN.R-project.org/package=mvabund>>
- William, Samuel M, et al. “Elk as a Potential Host for Meningeal Worm: Implications for Translocation.” *JSTOR*, vol. 56, Oct. 1992, pp. 629–639., doi:10.2307/3809453.

Appendix:

Figure 1: All gastropods included in graph between treatments and timing.

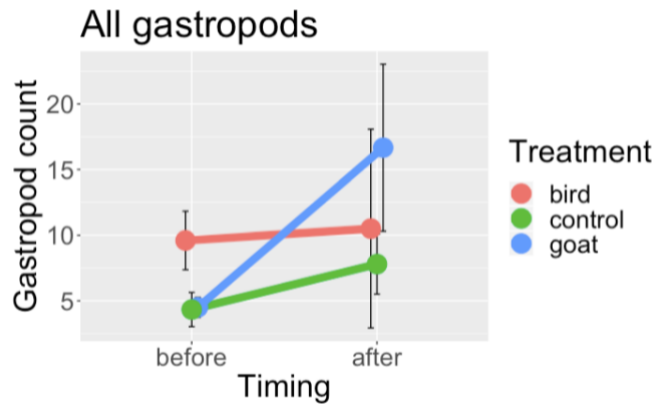


Figure 2: Density of known exotic slugs between treatment and timing.



Figure 3: Density of known intermediate hosts of *P. Tenuis* between treatments and timing.

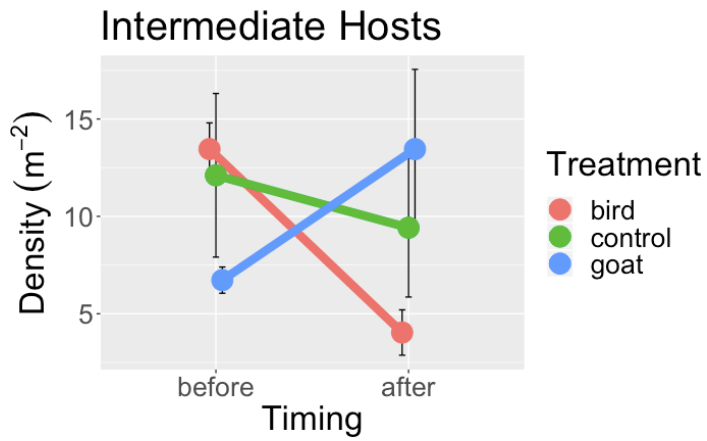


Figure 4: Shannon diversity measurements between treatments and timing.

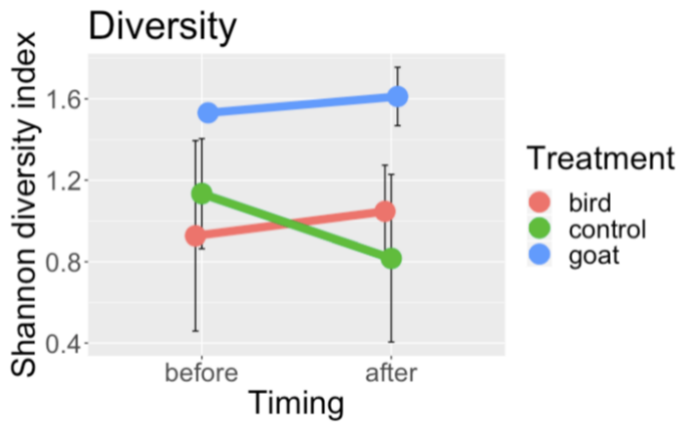


Figure 5: Simpson diversity measurements between treatments and timing.

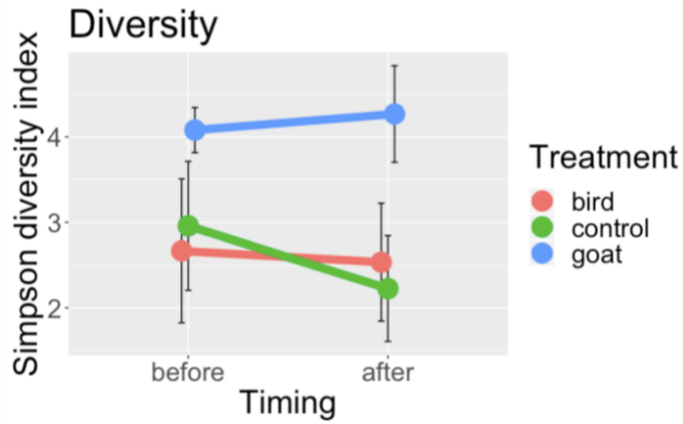


Figure 6: Richness diversity between treatments and timing.

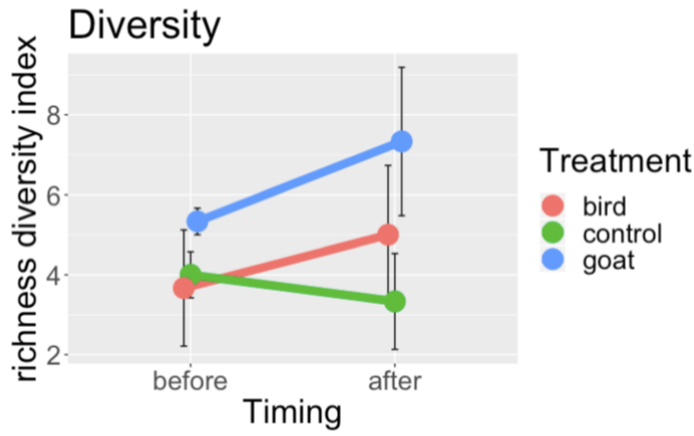


Figure 7: Points of each treatment and timing are grouped by hulls. The more overlap, the more similar the results.

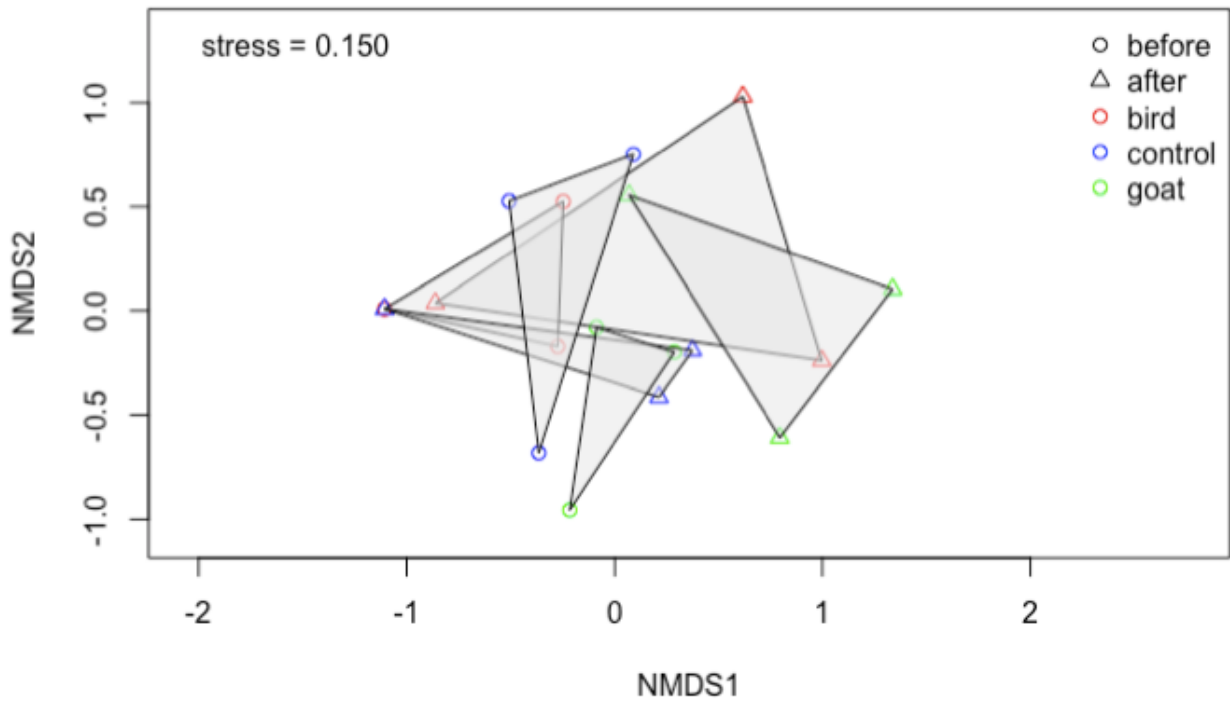


Figure 9: The length of the arrows symbolize how much of an effect that species had on the results. The direction symbolizes what treatment or timing they had an effect on.

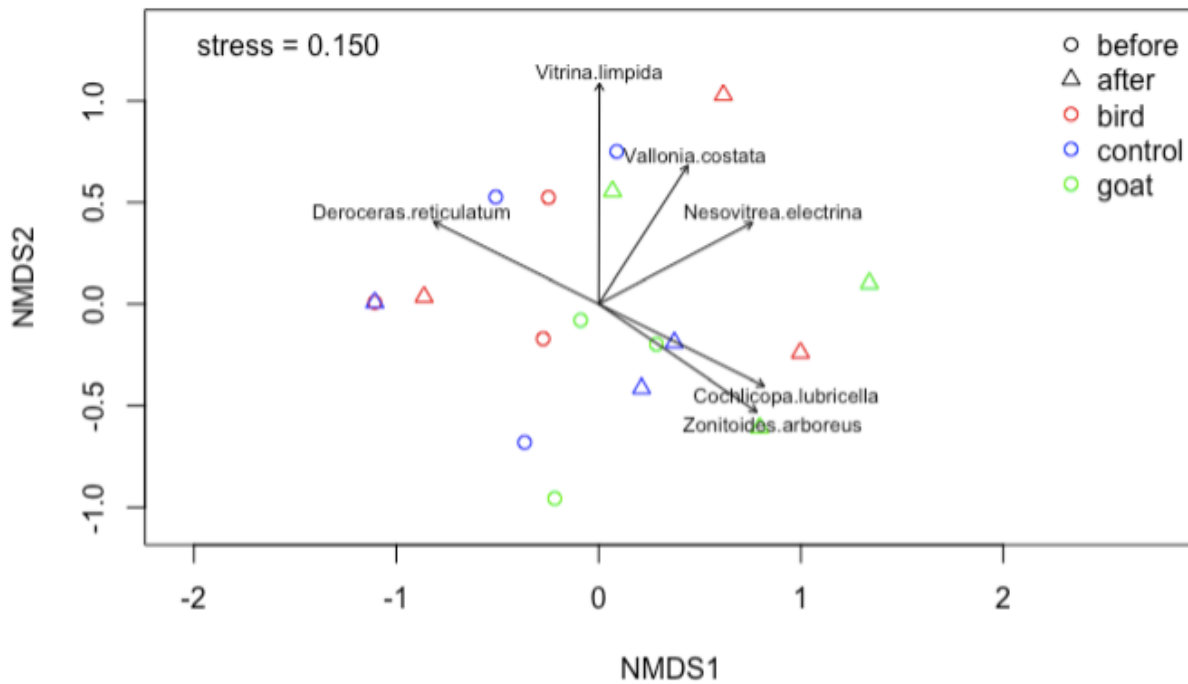


Figure 10: GLM for only intermediate hosts.

```
Call:
glm(formula = Number ~ Treatment * Timing, family = "poisson",
     data = h)

Deviance Residuals:
    Min       1Q   Median       3Q      Max
-1.5945 -0.3945 -0.1857  0.4638  1.4888

Coefficients:
              Estimate Std. Error z value Pr(>|z|)
(Intercept)    1.7918    0.2357   7.602 2.92e-14 ***
Treatmentgoat  -0.5878    0.3944  -1.490  0.1361
Treatmentbird   0.1054    0.3249   0.324  0.7457
Timingafter    -0.2513    0.3563  -0.705  0.4806
Treatmentgoat:Timingafter  0.9445    0.5263   1.795  0.0727 .
Treatmentbird:Timingafter -0.9527    0.5862  -1.625  0.1041
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

(Dispersion parameter for poisson family taken to be 1)

Null deviance: 26.363  on 17  degrees of freedom
Residual deviance: 13.867  on 12  degrees of freedom
AIC: 85.378

Number of Fisher Scoring iterations: 4
```

Figure 11: GLM of all gastropods.

```

Call:
glm(formula = Number ~ Treatment * Timing, family = "poisson",
    data = t)

Deviance Residuals:
    Min       1Q   Median       3Q      Max
-5.3474 -1.4589 -0.1405  1.1511  5.3619

Coefficients:
                Estimate Std. Error z value Pr(>|z|)
(Intercept)      2.15948   0.19612  11.011 <2e-16 ***
Treatmentgoat    0.03774   0.27477   0.137  0.8908
Treatmentbird    0.61310   0.24351   2.518  0.0118 *
Timingafter      0.40547   0.25318   1.601  0.1093
Treatmentgoat:Timingafter 0.90387   0.33338   2.711  0.0067 **
Treatmentbird:Timingafter -0.13353   0.31750  -0.421  0.6741
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

(Dispersion parameter for poisson family taken to be 1)

Null deviance: 186.37  on 17  degrees of freedom
Residual deviance: 115.23  on 12  degrees of freedom
AIC: 205.76

Number of Fisher Scoring iterations: 5

```

Figure 12: abundance of all species, using mvabund (Wang et al. 2020).

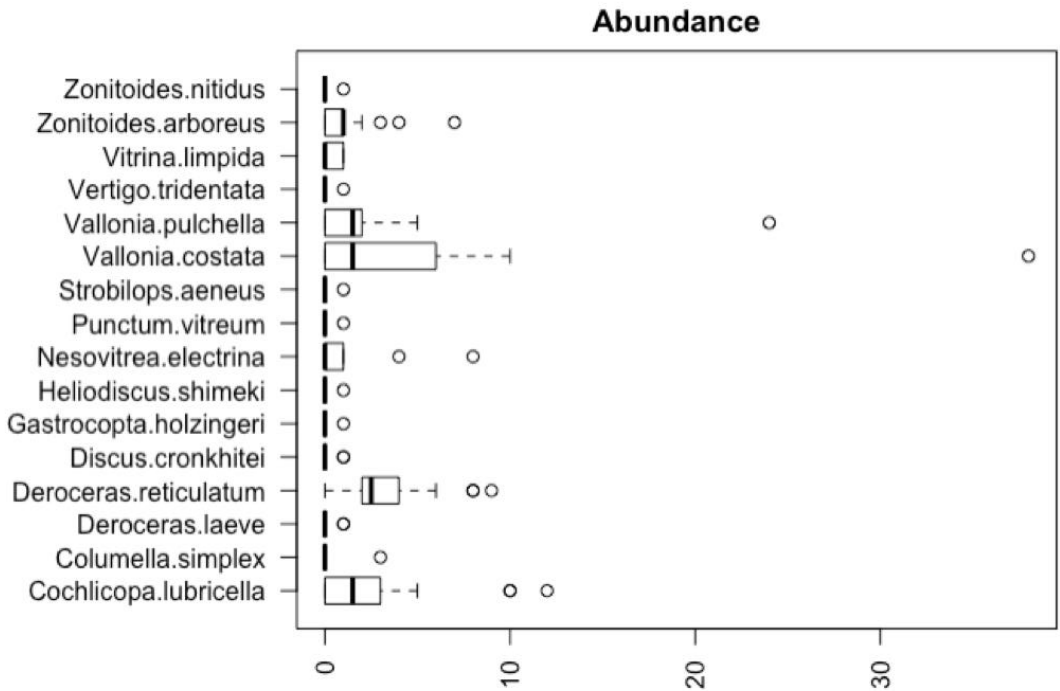


Figure 18: From *Discus cronkhite*, not *P. tenuis*.

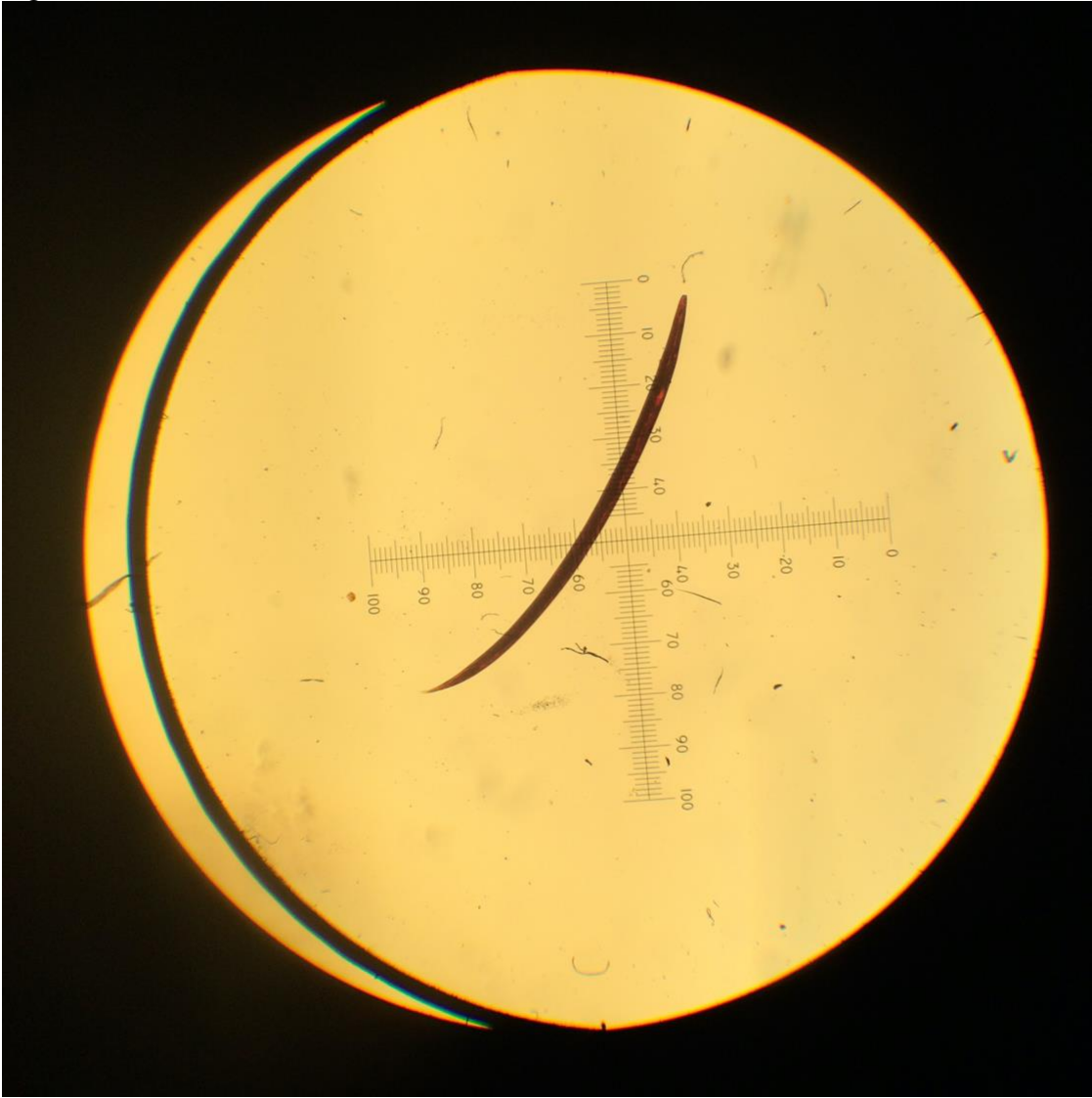


Figure 19: From *Deroceras reticulatum*, possibly *P. tenuis*.

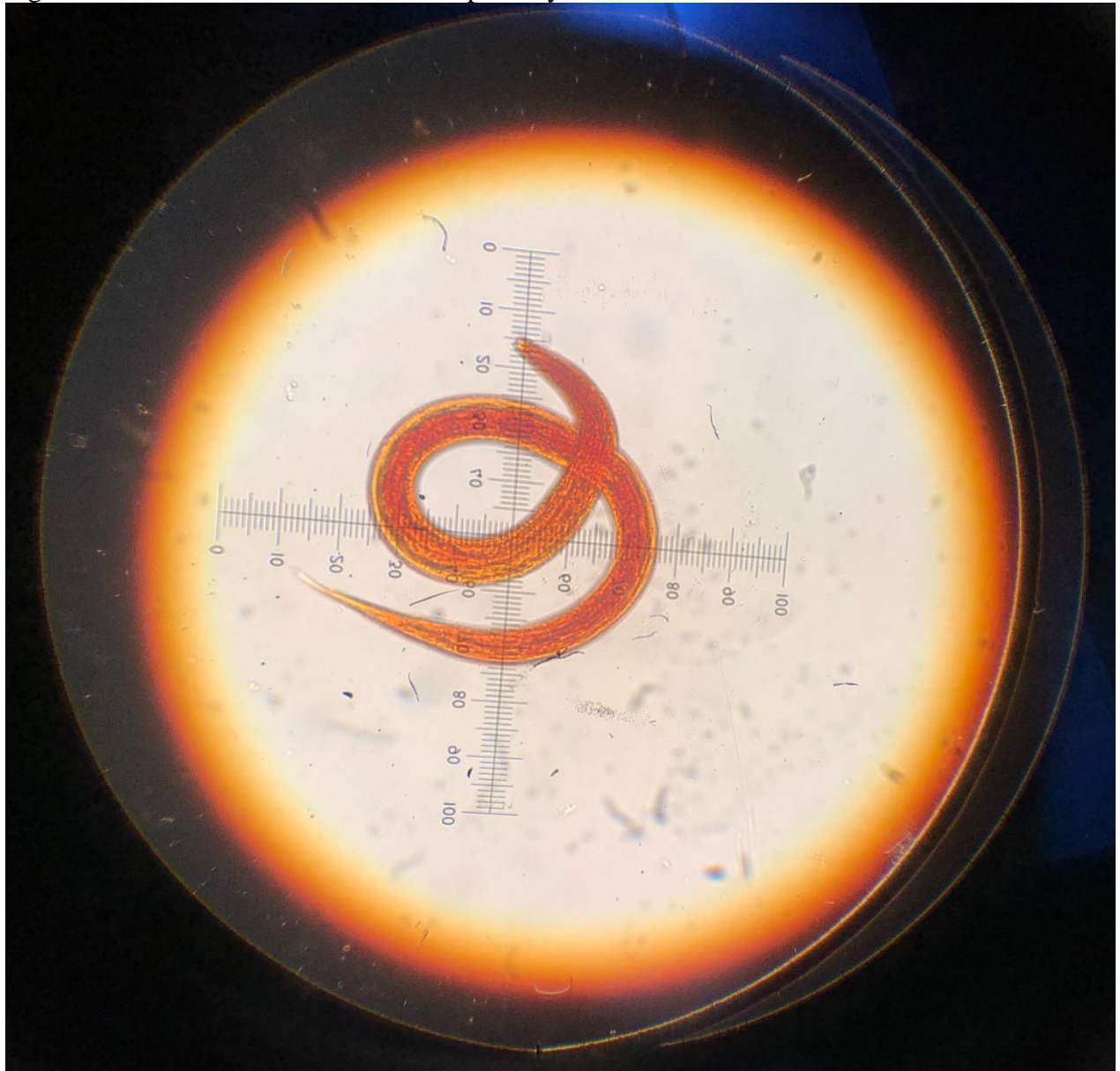


Figure 20: From *Cochlicopa lubricella*, not *P. tenuis*.

