Parasite surveillance in Prince Edward Island (PEI): screening previously frozen red fox lungs for *Crenosoma vulpis, Capillaria aerophilia* & *Angiostrongylus vasorum*

Savannah M. Christmas, Nina Germitsch, Nicole J. Guselle & Melanie Buote

Department of Pathology & Microbiology, Atlantic Veterinary College, University of Prince Edward Island, Prince Edward Island, Canada

Highlights

1) Retrospective nematode surveillance revealed high percentages (≥ 75%) of *Crenosoma vulpis* and *Capillaria aerophilia*-positive red foxes in PEI between the years 2012 and 2021.

2) *A. vasorum* was identified by morphological identification in fox samples from 2014, 2015 and 2021 when the previous earliest documented case in PEI was from 2018. Genetic sequencing is pending.

Introduction

1) *Crenosoma vulpis* (CV): Nematode parasite that inhabits the airways of dogs, foxes, and other wild carnivores[1]. It is common in foxes in Atlantic Canada. The definitive host is infected when they ingest an infected slug or snail[1].

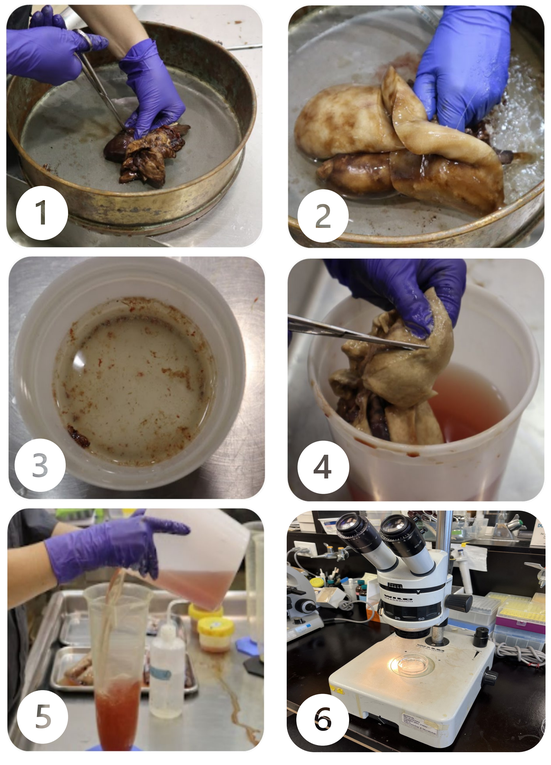
2) *Capillaria aerophilia* (CA): This nematode parasite resides in the airways of dogs, foxes, and other wild carnivores[1]. It has a worldwide distribution, and definitive hosts become infected directly or indirectly when they ingest an infected earthworm[1].

3) *Angiostrongylus vasorum* (AV): This nematode inhabits the blood vessels of wild canids[1]. First-stage larvae are passed in canine feces and are ingested by slugs or snails. Canids are infected after ingesting intermediate or paratenic hosts (e.g., frogs)[1].

Figure 1 A-D. Lungworms collected from red foxes. Figure 1A. CV (top), CA (middle) and AV (bottom) worms (12.5x mag.). Figure 1B. Caudal end of male CV (100x mag.). Figure 1C. Cranial end of female CA (100x mag). Figure 1D. Caudal end of male AV (100x mag).

Goals: This study aims to perform two screening techniques together (lung flush and dissection) to determine the prevalence of these common lungworms in a sample population of previously frozen PEI fox lungs.

Materials & Methods



1) Plucks (lungs with attached heart) were previously removed from red foxes and stored in the freezer at -80 °C.

2) After thawing, a tube is inserted into the heart's right atrium, and cold water is flushed through the blood vessels for 2 to 3 minutes.

3) Any parasites present in the airways will be flushed out of the lungs, trapped in a sieve and collected.

4) The lungs are then floated in a bucket of water. The arteries and airways are opened using scissors, allowing any parasites present to fall into the water.

5) The water is then transferred to a tube and allowed to sit for 30 minutes before the sediment at the bottom is collected.

6) The water from the flush and the sediment from the dissection are examined under a dissecting microscope. Any parasites found are organized by species and sex.

Results

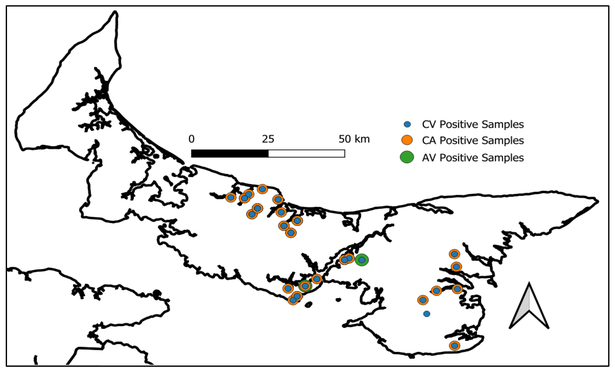


Figure 2. Map of PEI depicting where the positive red foxes were trapped.

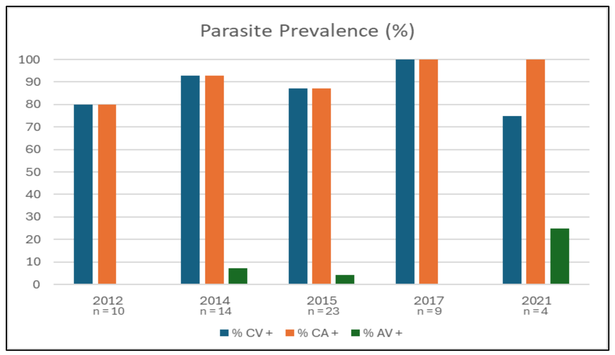


Figure 3. Yearly prevalence (%) of red foxes with CV, CA, and AV.

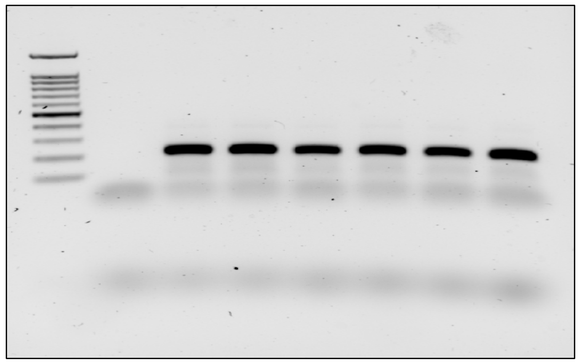


Figure 4. PCR test using AV4/AV5 primers that represents sufficient DNA extracted from possible *Angiostrongylus vasorum*. 1st Lane: DNA ladder (100 bp – 1,000 bp), 2nd Lane: negative control (no sample), Lanes 3 –8: nematode samples from two worms (A & B) taken from Fox #20.

Discussion & Conclusion

- In a study from 2005, fecal examination of 51 PEI foxes revealed that 78.4% tested positive for *C. vulpis* and 68.6% tested positive for *C. aerophilia*[2].
- As seen in Figure 3., performing lung flushes and dissection of 60 PEI foxes from between 2012 and 2021 revealed that 88.3% (n = 53) tested positive for *C.* vulpis and 90% (n = 54) tested positive for *C. aerophilia.*
- The higher prevalence in this study may indicate that infection prevalence has increased in PEI. However, sporadic shedding of nematode eggs in feces could account for such a difference in diagnostic tests.
- *A. vasorum* was first reported on PEI in 2022 in a study surveying wild canid feces[3]. Previously, the authors concluded that *A. vasorum* has been on the island since at least 2018[3].
- *A. vasorum* was identified by morphological identification in fox samples from 2014, 2015 and 2021. Genetic sequencing is pending.
- Future steps → continue lung flushes and dissections to have a larger sample size to confirm trends and statistical significance. In the next step, we intend to conduct an infectivity trial.

Literature Cited & Acknowledgements

1. Zajac, A. M. & Conboy, G. A. (2006). Veterinary Clinical Parasitology. Iowa, USA: Blackwell Publishing Professional.
2. Nevárez A et al. 2005. J Vet Diagn Invest. 17(5): 486-489.
3. Mahjoub H et al. 2022. CVJ 63(6): 637.

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