University of Prince Edward Island

Faculty of Veterinary Medicine
Summary of Dissertation

Submitted in Partial Fulfilment
of the Requirements for the

DEGREE OF DOCTOR OF PHILOSOPHY

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Alternative Metastrongyloid Parasite Transmission Routes from Gastropods to Definitive Hosts

Parasites within the superfamily Metastrongyloidea are nematodes that infect mammals. Generally, these parasites are found within the lungs of their definitive host. Metastrongyloid infections are significant in veterinary medicine, where effects on the host can range from subclinical infections to sudden death. Most metastrongyloids incorporate a gastropod intermediate host. Transmission between hosts represents critical stages of the parasites' life cycle. The first aim of this thesis was to determine the infectivity and retention of infectivity of gastropod-shed *Angiostrongylus vasorum* and *Crenosoma vulpis* L3 in dogs. Two "seed" dogs were exposed, resulting in patent infections. L1 collected from the seed dogs were used to infect slugs. Larval shedding was induced by anesthetizing the slugs in soda water and transferring them into warm water. Four naïve dogs were exposed to gastropod-shed L3, all resulting in patent infections. The second aim was to evaluate metastrongyloids in the terrestrial gastropod fauna on PEI. Terrestrial gastropods were collected and identified from sites on PEI during the summer of 2018 and 2019. Metastrongyloid L3 were collected from gastropod tissues and identified using DNA sequencing following PCR. *Arion fasciatus*-complex, *Deroceras laeve*, and *Cepaea hortensis* were gastropods found to have metastrongyloid infections. *Crenosoma vulpis*, *Filaroides martis*, and *Protostrongylus boughtoni* were the metastrongyloids identified. July had the highest average L3/gastropod and prevalence. The final aim was to assess the survival and decay rate of metastrongyloid L3 within a decomposing gastropod. Four experiments were undertaken using *A. vasorum* and *C. vulpis* L3. Living L3 were recovered up to three weeks post-mortem during all four experiments, with survival time decreasing and a maximum measured survival time of 6 weeks post-mortem. Migration of living L3 from the gastropod into the environment was low. The studies performed in this thesis have contributed to identifying potential routes of infection for metastrongyloid parasites.

Publications

Sokolov SG, Khrustalev AV, Greenwood SJ, Gray CN, **Robbins WT**, Jones MEB, Voropaeva EL, Kalmykov AP, Dzhamirzoev GS, Atopkin DM. 2023. Phylogenetic assessment of Apophallines (Digenea: Opisthorchiidae) with revision of *Apophallus donicus* Skrjabin & Lindrop, 1919 complex and some taxonomic propositions. Systematics and Biodiversity. 21(1):2189898

Robbins WT, Galeuzzi O, Graham K, Greenwood SJ, Jones, MEB, Buote M, Conboy GA. 2022. *Echinococcus multilocularis* infection in a red fox (*Vulpes vulpes*) on Prince Edward Island, Canada. The Canadian Veterinary Journal. 63(9):962-966.

Mahjoub HA, **Robbins WT**, Galeuzzi O, Graham KF, Jones MEB, Buote MA, Greenwood SJ, Conboy GA. 2022. First report of *Angiostrongylus vasorum* (French heartworm) in red foxes (*Vulpes vulpes*) on Prince Edward Island. The Canadian Veterinary Journal. 63(6):637-640.

Robbins W, Conboy G, Greenwood S, Schaper R. 2021. Infectivity of gastropod-shed third-stage larvae of *Angiostrongylus vasorum* and *Crenosoma vulpis* to dogs. Parasites & Vectors. 14(1):307

Lopez A, Aburto E, Jones K, **Robbins W**, Conboy G. 2016. *Eucoleus boehmi* infection in the nasal conchae and paranasal sinuses of red fox (*Vulpes vulpes*) on Prince Edward Island, Canada. Journal of Wildlife Diseases. 52(2):279-285.

Presentations

Robbins W, Yu J, Stryhn H, Greenwood S, Conboy G. Survival of Metastrongyloid Third-Stage Larvae in Experimentally-Infected *Limax maximus* Slugs Post-Mortem. Atlantic Canadian Association of Parasitologists (ACAP) Annual Meeting, Pictou, NS, Canada. November 25-27, 2022. (Regional Meeting, Oral)

Robbins W. Metastrongyloid L3 Survival in Deceased *Limax maximus* Slugs. Atlantic Canadian Association of Parasitologists' (ACAP) Annual Meeting, Virtual, October 23, 2021. (Regional Meeting, Oral)

Robbins W, Greenwood S, Conboy G. Preliminary Investigations into the Transmission and Survival Capabilities of Metastrongyloid L3. Atlantic Canadian Association of Parasitologists' (ACAP) Annual Meeting, Virtual, October 24, 2020. (Regional Meeting, Oral)

Robbins W, Greenwood S, Egers A, Schaper R, Conboy G. Snailed it! A Gastropod Intermediate Host Survey of Metastrongyloid Helminths on PEI. Atlantic Canadian Association of Parasitologists' (ACAP) Annual Meeting, Pictou, NS, Canada. October 25-27, 2019. (Regional Meeting, Oral)

Robbins W, Greenwood S, Rogers L, Conboy G, Schaper R. Infectivity of Shed *Angiostrongylus vasorum* & *Crenosoma vulpis* L3 to Dogs. 27th Conference of the World Association for the Advancement of Veterinary Parasitology (WAAVP), Madison, Wisconsin, United States of America. July 7-11, 2019. (International Meeting, Oral)

Robbins W, Greenwood S, Rogers L, Schaper R, Conboy G. Infectivity of Shed *Angiostrongylus vasorum* & *Crenosoma vulpis* L3 to Dogs. Graduate Studies and Research Days (GSR Days), UPEI, Charlottetown, Prince Edward Island, Canada. May 15-17, 2019. (University Meeting, Oral, Second-Best Presentation Award)

Robbins W, Greenwood S, Egers A, Buote M, Jones M, McRuer D, Conboy G. Infectivity of Spontaneously-Shed *Angiostrongylus vasorum* L3 to Dogs. Atlantic Canadian Association of Parasitologists (ACAP), Pictou, NS, Canada. October 19-21, 2018. (Regional Meeting, Oral)

Robbins W. A survey of the parasites of urban red foxes (*Vulpes vulpes*) in Charlottetown, Prince Edward Island, Canada based on fecal analysis. Science Graduate Studies Research Day, UPEI, Charlottetown, Prince Edward Island, Canada, June 23, 2014. (University Meeting, Oral)

Robbins W. A survey of the parasites of urban red foxes (*Vulpes vulpes*) in Charlottetown, Prince Edward Island, Canada based on fecal analysis. American Association of Veterinary Parasitologists (AAVP.) Annual Meeting, Denver, CO, USA. July 27, 2014. (International Meeting, Oral, Presentation Honorable Mention Award)

Robbins W. A survey of the parasites of urban red foxes (*Vulpes vulpes*) in Charlottetown, Prince Edward Island, Canada based on fecal analysis. Atlantic Canada Association of Parasitologists (ACAP), Pictou, NS, Canada. October 22, 2014. (Regional Meeting, Oral)

Biographical Data

Will Robbins was born and raised in Windsor, Ontario. He attended the University of Prince Edward Island (UPEI) where he obtained a Bachelor of Science degree (Biology) in 2013. During his undergraduate degree he performed summer research on green crabs (*Carcinus maenas*) under the supervision of Dr. Pedro Quijon. He also participated on an annual Department of Fisheries and Oceans (DFO) bottom trawl survey of the southern Gulf of St. Lawrence. In his last year of undergraduate he performed a senior undergraduate research project on urban red foxes (*Vulpes vulpes*) under the supervision of Dr. Marina Silva-Opps. From 2013-2018 he continued research on red foxes on PEI, where he received a Master's of Science under the supervision of Drs. Marina Silva-Opps and Dr. Sheldon Opps. Immediately following completion of his Master's degree, Will began his Doctor of Philosophy degree under the supervision of Drs. Spencer Greenwood and Gary Conboy in the Department of Biomedical Sciences at the Atlantic Veterinary College (AVC). In 2021, Will also began and is working towards his Doctor of Veterinary Medicine.

Will has instructed and assisted with laboratory sections in various courses in the Department of Biology at UPEI, including General Ecology, Vertebrate Zoology, Conservation Genetics, Biogeography and Macroecology, Biodiversity and Conservation Genetics, and Mammalogy. He has assisted with the Veterinary Parasitology laboratory at the AVC. He has also mentored 15 AVC Summer Research and Leadership (SRLP) students.

Will married Andrea in 2016. They welcomed their first son Liam in March, 2021 and their second son Alex in July, 2023. They also have two dogs – Kade (8 y) and Luna (8 mo).

Awards Received

Atlantic Veterinary College (AVC) Dean's Honor Roll (2021 – 2022 Academic Year), Atlantic Veterinary College Fall Recognition Night, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, PE, Canada, November 9, 2022

The Boehringer-Ingelheim Award in Parasitology, Atlantic Veterinary College Fall Recognition Night, Atlantic Veterinary College, University of Prince Edward Island, Charlottetown, PE, Canada. November 9, 2022.

Student Travel Award. 27th Conference of the World Association for the Advancement of Veterinary Parasitology (WAAVP), Madison, WI, USA. July 2019.

Second-Best Presentation Award. UPEI Graduate Studies and Research Days, Charlottetown, PE, Canada. May 15-17, 2019.

Student Presentation Honorable Mention Award. American Association of Veterinary Parasitologists' Annual Meeting. Denver, CO, USA. July 27, 2014

AAVP Young Investigator Travel Grant. American Association of Veterinary Parasitologists. July 2014.

Professor Earl Wonnacott Prize. The University of Prince Edward Island. February 2014.