

Studying American Water Shrew in the Peace with eDNA Methods



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Executive Summary

The American water shrew (*Sorex palustris*) is a comparably large, blackish-gray semiaquatic shrew species that forages for food in small streams and wetlands in the boreal forests and montane regions of northern, western, and central Canada. In British Columbia, *S. palustris* is listed as a species of special concern, and it has been identified as a priority species in the Fish and Wildlife Compensation Program's (FWCP's) Riparian & Wetlands Action Plan.

Considerable uncertainties exist with regards to the biology, distribution, and phylogenetic history of *S. palustris*, complicating the accurate assessment of limiting factors and the effective subsequent implementation of conservation actions for this listed species.

The subject project has been completed under the FWCP's 2020 Peace Region Action Plans. Specifically, the project has been completed under the Peace Region Riparian & Wetlands Action Plan Sub-Objective 4 and Action 7:

- Sub-Objective 4: maximize the population viability of priority riparian and wetland species,
- Action 7: research priority species to identify limiting factors (FWCP 2021).

This seed project includes a literature review of the existing species data and evaluates the feasibility, costs, and limitations of using eDNA to aid in determining this blue-listed shrew's range, distribution, and potential population threats in the Peace Region of BC.

eDNA technologies may represent a cost-effective, low-impact method to implement comprehensive inventory studies of cryptic species. However, the effective use of eDNA technology is predicated on the development of a species-specific assay for *S. palustris* based on nuclear DNA, which requires the acquisition of additional voucher specimens from northern BC.

This report provides a summary of the literature review on the biology of *S. palustris* and potential inventory methods to support future inventory work, as well as recommendations for the development of a larger project to support the management of this species of conservation concern.

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Table of Revisions

Revision No.	Date	Reason/Type of Revision
Draft R0	March 31, 2022	Draft report issued for FWCP review
R0	April 20, 2022	Incorporated FWCP review results, R0 report issued

1 Introduction

The Fish and Wildlife Compensation Program (FWCP), a partnership between BC Hydro, the Province of British Columbia (BC), Fisheries and Oceans Canada, First Nations, and Public Stakeholders, provides annual funding to projects aimed at conserving and enhancing fish and wildlife in watersheds impacted by existing BC Hydro dams. Seed Grant funding was available for 2020-2021 projects to allow individuals and organizations the opportunity to research large project application feasibility.

The FWCP Peace Project Area contains a wide variety of unique aquatic habitats that fulfill important ecosystem functions for a broad range of species, including riparian and wetland areas that may provide habitat for the blue-listed the American water shrew (*Sorex palustris*). Very little is currently understood about the *S. palustris*'s range, distribution, and population size in the FWCP Peace Region.

A desktop review utilized literature available to obtain information on the target species to gain a better understanding of their current status and population trends. Additionally, this project aimed to assess the feasibility of applying environmental DNA (eDNA) methods to help determine the presence and the distribution of *S. palustris* in the region.

2 Goals and Objectives

The subject project was completed under the FWCP's Peace Region Action Plans. More specifically, the project was completed under the Peace Region Riparian & Wetlands Action Plan Sub-Objective 4 and Action 7:

- Sub-Objective 4: maximize the population viability of priority riparian and wetland species,
- Action 7: research priority species to identify limiting factors (FWCP 2021).

This seed project includes a literature review of the existing species data and evaluates the feasibility, costs, and limitations of using eDNA to aid in determining this blue-listed shrew's range, distribution, and potential population threats in the Peace Region of BC.

3 Methods

Blackbird's project team completed an extensive review of peer-reviewed literature pertaining to the biology of *S. palustris* and small mammal inventory techniques, including a review of applicable conservation databases (BC CDC 2021).

Similarly, we engaged with biologists across the Province of BC to gather information on past projects involving small mammals, eDNA studies targeting water shrews, and historic data on shrew distribution throughout western Canada.

4 Results and Outcomes

4.1 *Sorex Palustris*

4.1.1 *Species Information*

The American water shrew (*Sorex palustris*) is a large, blackish-gray semiaquatic shrew species that forages for food in small streams and wetlands (Beneski and Stinson 1987, Nagorsen 1996).

Recent genetic and morphometric research has resulted in a taxonomic split of what has historically been considered *S. palustris* into three distinct species – *S. navigator* (western water shrew), *S. albibarbis* (eastern water shrew), and *S. palustris* (American water shrew; O’Neill et al. 2005, Mycroft et al. 2011, Hope et al. 2014). Only *S. navigator* and *S. palustris* are known to occur in western Canada, with research indicating potential for mitochondrial introgression between these two closely related shrew species in their contact zone in northeastern BC (O’Neill et al. 2005, Himes and Kenagy 2010, Hope et al. 2014, Nagorsen and Panter 2018).

The following focuses on *S. palustris* as defined following the split, as listed in the “Revised checklist of North American mammals north of Mexico, 2014” (Bradley et al. 2014).

4.1.2 *Distribution*

S. palustris is believed to range across boreal regions from Manitoba to the Northwest Territories, while *S. navigator* ranges from Alaska and the Yukon Territory to the southwestern United States.

There are only two confirmed historical museum specimen of *S. palustris* that originated in the Province of BC: one from Tupper near Dawson Creek, BC and one from Kotcho Lake near Fort Nelson, BC (D. Nagorsen, email message, February 11, 2021). No preserved tissues are available from the two historical specimens of *S. palustris* recorded in BC (Nagorsen and Panter 2018).

The western distribution limits of *S. palustris* in the northeast of BC and its contact zone with *S. navigator* are not well understood. The distributions of these two species are likely parapatric with a contact zone at the transition of the interior plains to the foothills of the northern Rocky Mountains (D. Nagorsen, email message, February 11, 2021). Uncertainties in the current known distribution reflect limited sampling effort to date.

4.1.3 *Habitat*

S. palustris is a habitat specialist that primarily utilizes the riparian habitat of small, shallow, cold streams with thick overhanging riparian growth (BC CDC 1996). First- and second-order streams are generally preferred (Kinsella 1967, BC CDC 1996). *S. palustris* has also been observed around lakes, ponds, wetlands, and other lentic habitats, though it appears to prefer lotic riparian systems (Kinsella 1967).

Availability of adequate cover in the form of overhanging banks, vegetation, boulders, tree roots, logs, etc. appears to play a large part in habitat selection by *S. palustris* (BC CDC 1996). Cover such as coarse woody debris is believed to be important in the establishment of suitable microhabitats for the purposes of nesting and foraging (Gustafson 2018). Decaying wood serves to retain environmental moisture, offering secure microclimatic conditions where there is abundant prey such as invertebrate species and plant foods (Beneski and Stinson 1987).

We were unable to find any structured studies conducted in the Province of BC on population trends, biology and habitat use, or on the movements of individuals of *S. palustris*.

4.1.4 Biology

S. palustris are semi-aquatic invertivores that hunt aquatic invertebrates and small fish under as well as on top of water. Diet analyses by Dunstone and Gorman (1998) suggest that *S. palustris* catches 50-90 % of its food in aquatic habitats.

Water shrews use a combination of olfaction and somatosensation (mediated by whiskers) to detect and pursue prey underwater (Catania et al. 2008). Their reliance on olfactory senses and somatosensation allow these nocturnal hunters to locate their prey without eyesight (Catania et al. 2008).

Water shrews, with their high metabolic rates, need to consume at least their weight in food every day (Conaway 1952, Sorenson 1962). In the wild they seem unable to store significant body fat and can die of starvation within a few hours. When a surplus of food is available, it is often hoarded, the shrew sometimes defecating on it to keep other shrews away.

S. palustris will reconstruct old nests or build entirely new ones, typically with dry vegetation in tunnels or under hollow logs. The nests built are approximately 8 cm in diameter (Nagorsen 1996).

S. palustris breed in late winter and early spring (February – March), with gestation lasting approximately 3 weeks (Conaway 1952). Litter sizes can range from 3-10 offspring with the average being 6; they can produce 2-3 litters per year under optimum conditions (Wilson and Ruff 1999). Most female water shrews will not reproduce until after their first winter (Beneski and Stinson 1987). *S. palustris* is known to have a relatively short lifespan of approximately 18 months (Nagorsen 1996, Wilson and Ruff 1999).

To attract a mate, the water shrew will take advantage of their well-established olfactory senses by emitting a strong, sometimes nauseating odor (Sorenson 1962). Water shrews are aggressive, and fighting is not uncommon. Sorenson (1962) found that male and female *S. palustris* were equally prone to fighting and no dominance hierarchy was evident. Fights are typically territorial in nature, which makes them solitary outside of the breeding season (Nagorsen 1996, Wilson and Ruff 1999).

S. palustris is an adept aquatic predator capable of sustaining dives from 31 to 47 seconds before exhausting themselves (Beneski and Stinson 1987). While diving the shrew propels itself using all four feet moving in a walking motion. Their hind feet are large and fibriated, providing maximum propulsion while swimming (Beneski and Stinson 1987). Shrews forage underwater by pressing their elongated snouts into the substrate. During a dive, the shrew is surrounded by a silvery layer of air trapped within the fur that increases buoyancy (Beneski and Stinson 1987). Throughout the winter, *S. palustris* will continue to use aquatic habitats, swimming beneath the ice (Beneski and Stinson 1987).

S. palustris, like most shrews, do not undergo prolonged torpor due to their significant metabolic energy requirements. They do, however, have enzymes that function well at low temperatures, which may facilitate daily torpor (Beneski and Stinson 1987). They have two distinct periods of major activity being reported as sunset to approximately 4 hours after, and just before sunrise (BC CDC 1996). Typically, the shrew will alternate between 30-minutes of activity and hour-long naps (torpor) (Sorenson 1962).

4.1.5 Population Sizes and Trends

No data are available. Because these shrews are habitat specialists, populations may be under threat of decline as suitable riparian habitat is modified, fragmented, or degraded.

4.1.6 Status Designation

S. palustris is currently blue-listed in BC (S2S4), with the global population classified as secure (G5). The provincial designation may be related to the absence of supporting population and distribution data, while the global classification reflects its widespread distribution across the boreal forests of Canada.

4.2 Inventory Methods for Small Mammals

4.2.1 Traditional Inventory Methods

Traditionally, shrew studies that require capture have used a variety of traps, including snap (kill), Sherman (live), pitfall (kill or live), and Longworth (live) traps (Brown 1967, BC RISC 1998). As a result of their semiaquatic lifestyle, water shrews can also be captured using minnow traps (kill or live; Welstead and Vennesland 2006, Craig and Vennesland 2007).

Ethical considerations should preclude the use of kill-traps in most ecological studies (particularly where the focal species is of conservation concern), and the following will focus on live trapping efforts.

Trappability of water shrew species depends on a variety of factors, including temperature, precipitation, moonlight, interaction between species and individuals within a species, as well as interactions between these factors. One of the main factors affecting trappability that researchers can influence, is the type of trap used.

Past studies, though limited in extent, have shown Sherman and Longworth traps to be relatively ineffective in capturing *S. palustris* (Stromgren 2008). During traps targeting shrew species, pitfall trap arrays, sometimes combined with drift fence funnels, appeared capable of capturing a wider range of species and more diverse individuals within a species than Sherman and Longworth traps (Stromgren 2008).

Live capture programs require careful study and procedural design to manage risks to individuals of the target taxa. Even under good conditions, mortality rates in trapping programs frequently exceed 25 % (Stromgren 2008, Ryckman 2020).

S. palustris' high metabolism and incessant need to eat high-caloric food drives most of its behavior and, together with an overall lack of understanding of its distribution and life habits, creates significant difficulties when attempting to study its life habitats (e.g., through mark-recapture programs). The addition of appropriate bait has been shown to significantly decrease trap mortality of shrews and is considered the standard for live trapping program for water shrews (BC RISC 1998, Do et al. 2013).

These described traditional capture methods for shrews suffer from low capture rates and high trap mortalities (Stromgren 2008). More recently used methods include scat tubes and camera traps. Camera traps modified with lenses for close focal distances have shown high detection rates for shrews, while baited scat tubes have been used successfully in the context of eDNA surveys for *S. bendrii* (Dykshoorn 2004, Tennant et al. 2020, Delisle et al. 2021).

4.2.2 eDNA

eDNA monitoring and assessment tools have experienced a significant increase in applications and the development of standardized methods for the collection, analysis, and interpretation of replicable data in applied biology applications (Thomsen and Willerslev 2015). eDNA techniques promise cost-effective, less invasive presence/not-detected surveys with relatively high detection probabilities of cryptic species compared to traditional inventory methods (Biggs et al. 2014, Goldberg et al. 2015).

eDNA methods are predicated on the fact that species exogenously shed their genetic material into their environment as they complete their life processes, and this exogenous DNA may become suspended in aquatic ecosystems (Pilliod et al. 2013, Maruyama et al. 2014). Genetic material can be collected through filtration of water sampled from the target taxa's suspected habitat or through swabs when baited scat tubes are used.

eDNA is characterised by a complex mixture of nuclear and mitochondrial DNA and can be intracellular (from living cells) or extracellular. Most eDNA analyses developed to date target short fragments of mitochondrial DNA (mtDNA), as every eukaryotic cell usually has two copies of nuclear DNA and hundreds or thousands of copies of mitochondrial DNA. The comparably large number of mtDNA copies allows the optimization of species or taxa from environmental samples.

However, mtDNA is generally maternally inherited, which precludes the identification of hybrids. eDNA as a method to detect *S. palustris* is subject to limitations based on the known mitochondrial introgression between *S. navigator* and *S. palustris*, which would require use of a nuclear marker.

5 Discussion & Recommendations

The limited body of knowledge on the distribution, biology, and population trends of *S. palustris* and the recent split of the *S. palustris* complex in western Canada into *S. palustris* and *S. navigator* severely restricts the effective assessment and management of this species of potential special concern. The Peace FWCP region, due to its significant overlap with the suspected contact zone between the two species, provides a unique opportunity to expand the body of knowledge on *S. palustris* and ultimately inform future inventory, management, and habitat restoration projects that may benefit this species.

S. palustris and *S. navigator* are not reliably distinguishable based on external features. As a result, an accurate taxonomic assignment of an individual requires either morphometric measurements or genotyping, making the identification of live captures in hand impossible (Nagorsen et al. 2018).

eDNA methods, though potentially challenging to apply to semiaquatic species, may hold promise in determining *S. palustris*'s distributional limits and extent of sympatry among contemporary populations of water shrews in western Canada (Yonezawa 2020).

The potential use of eDNA presents a less invasive sampling technique, which is important when sampling for species as sensitive as *S. palustris*. The availability of a species-specific assay that targets a small portion of the genome and reliably helps identify the presence of *S. palustris* is key to the successful implementation of an inventory program based on eDNA technology (Farrington and Lance 2014). Mitochondrial introgression between parapatric populations of water shrews will require the use of a nuclear marker, the development of which will require additional voucher specimens from northern BC.

Water shrews are known to be caught as bycatch during fisheries inventory studies (Welstead and Vennesland 2006, Craig and Vennesland 2007). It is recommended that researchers work with provincial fisheries biologists to retain shrew specimens accidentally caught during otherwise unrelated fisheries research work (e.g., through the inclusion of a recommendation or condition on applicable regulatory permits under the *BC Angling and Scientific Collection Regulation*).

In addition, future research should consider the inclusion of a baited camera trap component (e.g., as a reconnaissance survey) to aid in the selection of sampling sites and increase the probability of trapping success.

6 Acknowledgements

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