Proceedings of the Nova Scotian Institute of Science (2025)
Volume 54, Part 1, pp. 135-166

USING RAPID AND REPEATABLE SIDE SCAN SONAR METHODS FOR A SECOND ASSESSMENT OF THE SHORTNOSE STURGEON (*ACIPENSER BREVIROSTRUM*) POPULATION IN THE SAINT JOHN RIVER, NEW BRUNSWICK, CANADA

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ABSTRACT

Population estimates are a key component of fisheries management, particularly when assessing species of concern. However, the time and effort required to conduct those estimates logistically limits their frequency. To facilitate assessment of Shortnose Sturgeon (*Acipenser brevirostrum*; SNS) which are a species of concern in the Saint John River, New Brunswick, Canada, a combined side-scan sonar and acoustic telemetry-based method was employed to enumerate SNS within high density winter aggregations. During this study 12,005 SNS were enumerated in one main winter aggregation and 2,186 SNS were counted within a second in the Kennebecasis Bay. Winter residency patterns determined from acoustic tracking of 18 tagged SNS over 8 years (2015/16-2022/23) indicated that these two aggregations represented on average 74.3% of the overall population suggesting that the total Saint John River population was ~19,100 SNS > 40 cm FL in winter 2022/23. Although the development of more in depth, robust, and repeated assessments are needed to verify this estimate of abundance and size classes, we conclude that the abundance of SNS in the Saint

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John River has probably remained stable since the earliest population estimate completed in 1977.

Keywords: Winter Aggregation, Machine Learning, Acoustic Telemetry, Population Monitoring, Threatened Species

INTRODUCTION

Sturgeons (family *Acipenseridae*) are long-lived fishes (Sulak and Randall 2002) native to rivers, lakes, and coastlines across all continents of the Northern Hemisphere (Haxon and Cano 2016), but throughout this range nearly all populations are in decline (Lenhardt *et al.* 2006). The life history strategy of sturgeons which includes slow growth, late maturation, and high survival and fecundity (Tripp *et al.* 2009) has weathered millennia of natural environmental upheavals (Choudhury and Dick 1998). However, when faced with a world of rapidly evolving and compounding anthropogenic challenges, this strategy may no longer be advantageous. Globally, Sturgeons are threatened by dams which incur passage mortality and fragment habitats (Huang and Wang 2018), pollution that degrades water quality (Blevins 2011); affects reproduction (Feist *et al.* 2005); and reduces juvenile survival (Hummel *et al.* 2022), climate change that warms global waters (Lassalle *et al.* 2010), and overharvest which directly impacts spawner abundance (Quist *et al.* 2002, Chambers *et al.* 2012). As sturgeon populations worldwide trend towards low abundances, rates of decline become harder to measure as the species themselves become more difficult to observe. This challenge becomes particularly apparent when those species occur in large rivers and lakes. Without the ability to easily gather data, factors leading to population decline likewise become more difficult to mitigate and the time and costs associated with acquiring those necessary data can become a significant impediment to conservation.

The Saint John River (SJR), New Brunswick (NB), is the largest Canadian River east of the St. Lawrence, and the only river in Canada known to support a reproducing population of Shortnose Sturgeon (*Acipenser brevirostrum* Lesueur 1818; SNS). The occurrence of SNS in this system is likely due to the extensive, warm, mesohaline estuarine habitat enabling the amphidromous life history of this species (Miller 1972, Dadswell 1979, Fisheries and Oceans Canada 2016). In this system the habitats of Shortnose Sturgeon have been restricted by the construction of hydroelectric dams. The largest and most downstream of these barriers is the Mactaquac Dam which is located immediately upstream from an important spawning location (Usvyatsov *et al.* 2012) and forms a definitive barrier to upstream movement. Additional potential threats to the species in the SJR include industrial pollution (Dadswell 1979), residual dichlorodiphenyldichloroethylene (DDE) from past forest spraying with dichlorodiphenyltrichloroethane (DDT; Elson 1967), elevated total mercury (Dadswell 1975, Andrews *et al.* 2023), and possibly invasive species (Bruce *et al.* 2019, Zelman *et al.* 2023). Catch and release angling could also have an impact (Struthers *et al.* 2018), but those potential effects are unknown. In 1980, 2005, and 2015 the Committee on the Status of Endangered Wildlife in Canada (COSEWIC) designated the genetically distinct SJR SNS population (SSSRT 2010, Wirgin *et al.* 2010, King *et al.* 2014) as "Special Concern" (Dadswell 1980, Dadswell 1984, COSEWIC 2005, 2015). This designation was not based on current population trends which were unavailable. Rather, it was predicated upon the suspected occurrence of a single spawning location (Li *et al.* 2007, Usvyatsov *et al.* 2012) and the ensuing vulnerability imposed by the limited distribution of SNS in Canada. The COSEWIC designation was followed by an official listing under Schedule 1 of the Species at Risk Act (SARA) in 2009. This action prompted the creation of a management plan directing federal conservation, management, and monitoring efforts for the species (Fisheries and Oceans Canada 2015, 2016).

The most recent management plan for SNS in Canada lists "estimates of abundance" as a top priority for ongoing monitoring and management (Fisheries and Oceans Canada 2016). Several estimates of abundance for the SNS population (or components of the population) in the SJR have been attempted over past decades of which the most comprehensive reported ~18,000 ± 5,400 individuals >50 cm FL (fork length) following an intensive 5-year mark recapture study spanning 1973-1977 (Dadswell 1979). Subsequent assessments in 2005, 2009 and 2011 used video surveys of a localized winter aggregation in the Kennebecasis branch of SJR (Fig 1) over three months (January to March) and predicted a site-specific population of 4,836, 3,852 and 5,222 in each of those years respectively (Li *et al.* 2007, Usvyatsov *et al.* 2012). Mark recapture estimates were also conducted in the Kennebecasis between 1998-2004 using fish captured during an annual angling derby. That study produced an estimate of 2,068 individuals in the Kennebecasis, but the 95% confidence interval ranged widely (801-11,277; COSEWIC 2015). Most recently in 2018, population abundance was estimated using side imaging sonar coupled with a 4-year acoustic tracking data set (Andrews *et al.* 2020). That study directly enumerated 12,284 SNS on side imaging sonar during a 1-day survey, an estimated 61.1% of the SJR population. From this finding a whole river population was estimated as 20,101 > ~40 cm FL (Andrews *et al.* 2020). This estimate was comparable to the adult population of 18,000 ± 5,400 >50 cm FL estimated by Dadswell (1979) and was the first attempt at a full river population estimate in over 40 years.

Sonar systems, both simple and sophisticated are being more commonly employed to conduct fish population and habitat surveys (Kaeser 2010, 2012, Flowers and Hightower 2013, 2015). Advances in sonar technology have tightened gaps in image quality and post

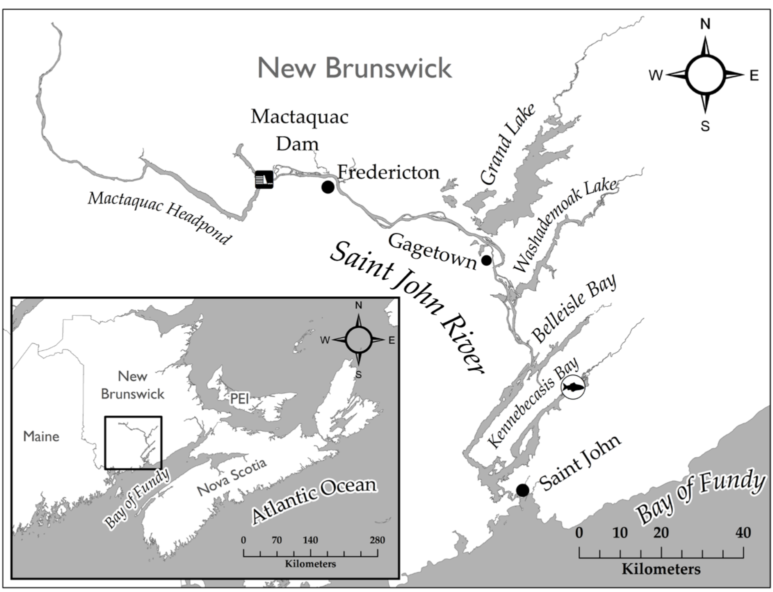

Fig 1 **Saint John River, New Brunswick, extending from its confluence with the Bay of Fundy at the City of Saint John upstream past Kennebecasis Bay, Belleisle Bay, Washademoak Lake and Grand Lake until meeting the Mactaquac Dam upstream from the City of Fredericton at river km 150. An aggregation of Shortnose Sturgeon is described to form in late fall and winter in the upstream portion of Kennebecasis Bay near the confluence of the Kennebecasis and Hammond rivers and is indicated by the fish symbol. Image reproduced from Andrews *et al.* (2020) with permission.**

processing capabilities between inexpensive consumer grade electronics (*e.g.,* Helminen *et al.* 2019) and professional commercial sonar systems (*e.g.,* Helminen and Linnansaari 2023). These advances have led to a transition from using inexpensive sonar systems as tools for depth and general fish habitat mapping (Buscombe 2017) to an effective means of surveying fish populations. While the most common sonar systems are dual beam and operated actively from a boat, some of the most effective fish sonar enumeration systems are situated such that the target species is guided past stationary multibeam sonars positioned in narrow rivers (Magowan *et al.* 2012, Helminen and Linnansaari 2023) or engineered fish passage structures (Grote *et al.* 2014). The most sophisticated of these systems incorporate artificial intelligence which can be trained to both identify species (Helminen *et al.* 2021) and rapidly enumerate targets in real time (Kandimalla *et al.* 2022, Connolly *et al.* 2022). Though broadly applicable, stationary sonar installments are most suited to monitoring anadromous species because spawning adults ascend rivers in large numbers within predictable seasons (Hughes & Hightower 2015), often passing both natural and man-made bottlenecks. The behaviour and migratory pathway of highly motivated anadromous species greatly facilitates this method.

Shortnose Sturgeon in the SJR, NB, conduct an upstream spawning migration to freshwater, but among individuals and sexes, these migrations have infrequent periodicity (Dadswell 1979, COSEWIC 2015). The size of the SJR itself (800 m-1 km wide) and lack of bottlenecks also make passive sonar monitoring for SNS impractical. Rather, exploration of winter habitats where SNS aggregate densely and remain in-situ for long periods of time provides a more practical solution (*e.g.,* Li *et al.* 2007). By surveying winter aggregations with boat mounted sonar, the limitations of fixed sonar can be mitigated, and its application reversed whereby a mobile sonar unit can be used to enumerate stationary fish (Andrews *et al.* 2020 see Flowers and Hightower 2013 and 2015 for a comparison to summer sonar surveys in a large river). This method has demonstrated potential in estimating river wide abundance when paired with telemetry-derived winter residency proportions of SJR SNS (Andrews *et al.* 2020). Given the recent applicability of sonar advances, the present study attempts to enumerate both the main winter aggregation (described by Andrews *et al.* 2020) and the Kennebecasis winter aggregation (described by Li *et al.* 2007 and Usvyatsov *et al.* 2012) of SNS in the SJR with consumer-grade side-scan sonar. The goal of this study is to provide a step forward in developing the necessary tools and methods for effective, rapid, and inexpensive monitoring of Canada's only population of SNS.

STUDY AREA

The Saint John River (Fig 1) is the largest river draining into the Bay of Fundy which it meets along the western shore of the bay at the City of Saint John, NB. Once free flowing for 673 km (Cunjak *et al.* 2011), the SJR is now obstructed by three large main-stem hydroelectric dams (Ruggles & Watt 1975). These barriers include 1) Grand Falls Dam located 350 km upstream from the river mouth constructed in 1931, 2) Beechwood Dam located 285 km upstream from the river mouth constructed in 1952, and 3) Mactaquac Dam constructed at river km 150 in 1968 (COSEWIC 2015). The Mactaquac Dam, which is the largest and most downstream passage barrier limits the upstream movements of most species including SNS. Downstream passage at Mactaquac Dam is facilitated only by transit through the turbines, spillways, or sluiceways when operational (Babin *et al.* 2020). Supervised upstream fish passage by means of a fish lift and trap and truck operation are facilitated for *Salmo salar* (Atlantic Salmon, L. 1758), limited quantities of Alewife (*Alosa pseudoharengus* Wilson, 1811), and Blueback Herring (*Alosa aestivalis* Mitchill, 1815), while all other species are manually filtered from the lift and returned downstream (Chateauvert *et al.* 2018). SNS have been captured during boat electrofishing surveys immediately downstream of Mactaquac Dam as recently as 2022 (R. Hill, Ph.D. student at UNB Fredericton pers. comm.). Though only a single Shortnose Sturgeon observed in 1968 (Smith 1979; the first year of operations at Mactaquac Dam) was ever recorded entering fish passage collection facilities (Meth 1972, Williamson 1974, Smith 1979, Ingram 1980, 1985, Jessop 1990, Hooper 1991, R. Beaumaster, Fisheries and Oceans Canada fish lift technician, pers. comm. 2016). Fish passage structures at Mactaquac Dam which were designed as a surface collection facility are inappropriate for the attraction and passage of SNS or other benthic species (*i.e.* American Eel; *Anguilla rostrata* Lesueur, 1821).

From the mouth of the SJR, tidal influence extends 130 km upstream to the city of Fredericton (Fig 1) with saltwater intrusion detectable ~70 km upriver to the village of Gagetown (Fig 1; Carter & Dadswell 1983). The lower reaches of the SJR are fed by four main-stem tributaries including Grand Lake, Washademoak Lake, Belleisle Bay, and Kennebecasis Bay which are subject to increasing degrees of tidal influence and proximity to the river mouth in that order. The upper portion of Kennebecasis Bay supports an extensive salt marsh (Hampton Marsh) at the mouth of the Kennebecasis River. This region contains a well documented winter aggregation of SNS (Li *et al.* 2007, Usvyatsov *et al.* 2012) that occupies a sandy thalweg 4.5-7 m deep near the confluence of the Kennebecasis and Hammond rivers. The Hampton Marsh itself is interspersed with tidal channels and pockets of various depth providing additional refugia for SNS in autumn and winter.

METHODS

Workflow

This study reproduced the methodology for a sonar-based population estimate of SNS in the SJR as described by Andrews *et al.* (2020) but expanded upon the number of surveyed areas to produce a more comprehensive estimate during winter 2022/2023. This updated study iteration surveyed the main aggregation described by Andrews *et al.* (2020) in addition a well-documented winter aggregation in the upper Kennebecasis Bay (Fig 1; Li *et al.* 2007, Usvyatsov *et al.* 2012). During surveys, parallel side-scan sonar transects were conducted with a Humminbird® (Johnson Outdoors, Racine, WI, United States) Helix 10 MEGA SI Gen 4 fish finder. These data were collated in Reefmaster® software (Reefmaster Software Ltd. Birdham, UK) to produce an image mosaic of each surveyed aggregation that were exported as mtbtiles for manipulation in QGIS and transfer to AcrPro. Within the GIS platform image pixels were classified either as "sturgeon" or "riverbed" using a supervised maximum likelihood model (*sMLC*) to produce an enumeration of "sturgeon" in each image mosaic. These estimates were further refined by calculating the minimum bounding geometry of each shape and applying an inclusion threshold based on size as per Andrews *et al.* (2020). The population counts across both surveyed locations were then compared to winter residency proportions determined from eight years of continuous acoustic tracking of 18 tagged SNS to create a population estimate for the entire SJR (Fig 2).

Side-scan data collection

Side-scan sonar surveys were conducted using a commercially available Humminbird® Helix fish finder and were recorded at a frequency of 1,275 kHz while scanning 30 m to either side of the survey vessel. Survey speed varied from ~3-9 km/h during transects depending on wind and current speed, and each survey was conducted as a series of parallel unidirectional tracks each saved as individual recorded files (Fig 3). Transects were started at one side and in parallel to the orientation of the sturgeon as observed on side-scan sonar to capture the fish in profile. Survey passes were then conducted in parallel, offset by ~25-30 m. This process was repeated until the entire aggregation had been covered and sturgeon ceased to appear on sonar. During surveys GPS (Global Positioning System) position, orientation and speed was continuously recorded by the fish finder. The positional difference between the head unit and the transducer, as well as the depth of the transducer below the water line were corrected prior to image mosaicking. Transects were driven manually and efforts were made to keep transects as straight and uniform as possible. Although wind and waves inevitably resulted in some yaw and sway in the trajectory of the vessel and thus movement of the transducer.

Side-scan sonar survey of winter aggregations

On Nov 24 and 25, 2022, the main aggregation was fully surveyed three times (two surveys on Nov 24, and one on Nov 25) with survey taking 4 to 5 hours of continuous scanning to complete. During scanning, wind speed ranged from 2 to 3.6 m/s though increased to >5 m/s in the afternoon of Nov 25th, removing the possibility to complete a 4th side-scan transect on that day. Surface water temperature during these surveys ranged from 1.2-1.4 °C, a temperature at which SNS form dense winter aggregations and remain relatively immobile (Andrews *et al.* 2020). Like the 2018 survey, side-scan passes over the main aggregation were completed during the outgoing tide as reversals in tide result in re-orientation and shuffling of the aggregation.

On Nov 27, 2022, three side-scan passes were conducted over the Kennebecasis aggregation during a single outgoing tide. Each of the three surveys took ~1 hour to complete. Wind speed at the time of

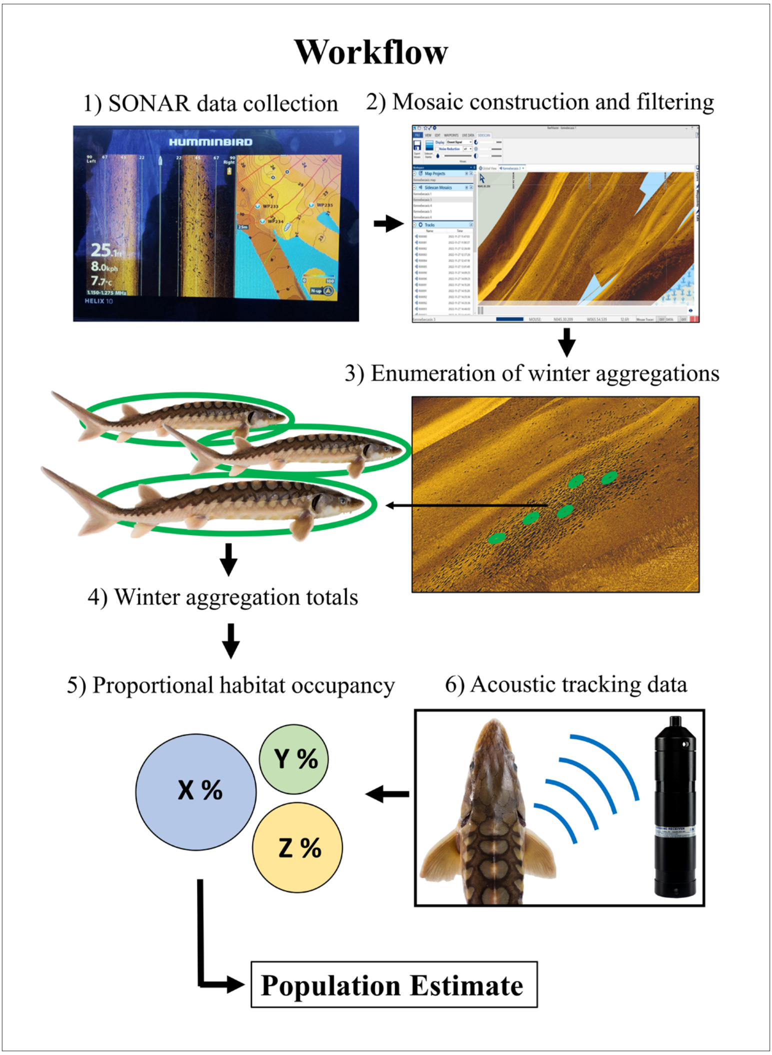

Fig 2 **Project workflow diagram following sequential steps including 1) collection of side-scan sonar data, 2) creation of side-scan mosaic mtbtiles file, 3) enumeration of scanned winter aggregations to create 4) totals within aggregations. These totals were then assessed in accordance to 5) winter habitat residency proportions derived from 6) an 8-year acoustic tracking data set from 18 tagged Shortnose Sturgeon (*Acipenser brevirostrum*) to calculate a final river-wide population estimate in the Saint John River, New Brunswick.**

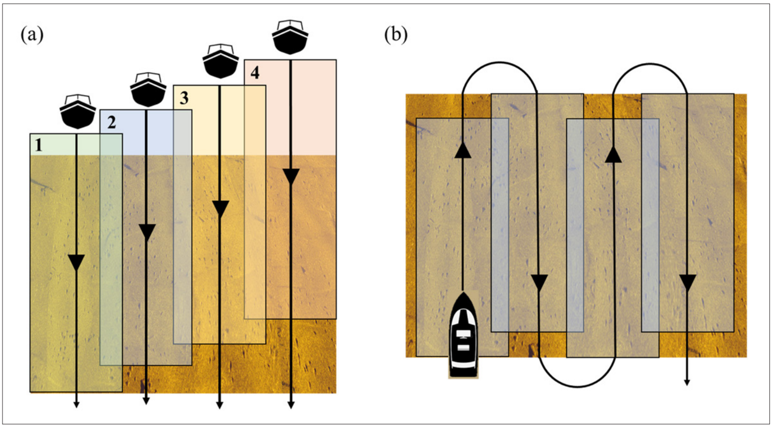

Fig 3 **Comparison of side-scan sonar track methods demonstrating (a) the method used in this study which involved separate evenly spaced parallel scans each saved as a separate sonar file, and (b) the method used by the prior study (Andrews *et al.* 2020) which involved a single continuous scan saved as a single sonar file where the scanned area was covered in two directions and later filtered to remove regions of maximum curvature.**

this survey ranged 1.5-2 m/s while surface water temperature was 0.8°C. At the time of the survey, SNS were observed to be very tightly aggregated, and this aggregation was scanned almost in its entirety in a single sonar pass. Additional surveys were also conducted along tidal channels adjacent to the Kennebecasis winter aggregation to count SNS not relating to the core group. These surveys were not comprehensive within the maze of available tidal channels of the Hampton Marsh, but three separate side-scan passes were completed for a small wintering assemblage located ~1.5 km away from the core group. No control summer surveys were conducted as part of this study because data for the main aggregation was collected by Andrews *et al.* (2020) and the Kennebecasis is a well documented winter aggregation (Li *et al.* 2007, Usvyatsov *et al.* 2012).

Side-scan sonar image mosaicking and filtering

During operation, the side-scan transducer produced multiple scan lines each second and simultaneously, the Humminbird® fish finder measured the GPS location, speed, and rotation of the boat which it records approximately 1-3 times per second. Using Reefmaster® software, the continuous scan lines recorded by the transducer were compiled to produce a 2D acoustic image or side-scan "mosaic". This mosaic was corrected using the "Blend Closest Display" setting in Reefmaster® which prioritizes the signal closest to the center of the side-scan transect and blends the tracks at the margins of the swath for a more seamless overlap between recorded transects. The final mosaic was then processed though 1x noise reduction and 100% autogain to equalize brightness across the image. In some cases, boat movement resulted in image distortion and stretch. These instances were noted, and while the visualized fish could still be counted, some could have been filtered from the estimates. Removal of individuals from the estimates could have occurred if those stretched images resulted in SNS appearing to exceed pre-set target length restrictions defined during image post processing (see image classification). Filtration of stretched images would have resulted in lower overall counts. Following image mosaicking data were transfer to ArcGIS as a pixel resolution of 7.5 cm.

Image classification

Maximum likelihood classification (sMLC) was used to classify objects in the sonar images. These objects were characterized as "Riverbed" or "Sturgeon". The initial training data were manually denoted by visually inspecting the sonar image and drawing polygons around objects that were either identified as riverbed or suspected sturgeon. These polygon features were then used to classify the images. All the data was processed in ArcMap 10.8 (ESRI, Redlands, CA, United States).

Following this classification, an additional filtering method was applied. This method is detailed in Andrews *et al.* (2020) and in brief entails calculating the "minimum bounding geometry" of the classified images meaning that an inclusion threshold was set using the length of the objects classified. Andrews *et al.* (2020) used an inclusion threshold for surgeon polygons of 20 cm < Potential SNS < 150 cm. The minimum limit served to remove bottom debris from classification as SNS, and the upper limit was the maximum length of SNS. During this survey we used a more conservative inclusion threshold of 40 cm < Potential Shortnose Sturgeon < 150 cm because 40 cm was the functional resolution of the sonar unit as described by Andrews *et al.* (2020).

A kappa coefficient (k) was calculated to assess the validity of the sMLC classification. Given the lack of actual observations, N = 105 manual classifications were demarked as riverbed (n = 40) and sturgeon (n = 65). We repeated this for each assessed site. These data were then used to calculate *k* and both analyses were completed in Excel (Microsoft 2023).

Our original goal was to classify and enumerate sturgeon captured within each collected sonar mosaic allowing for the reporting of variability and confidence limits between repeated estimates. However, some sonar passes were compromised by wind and wave action, and we elected to only analyse the cleanest mosaic taken at each study site for inclusion in our population estimate. Additional survey days were not possible in winter 2022 because both sites were accessed as late in the season as ice conditions would allow and wind conditions deteriorated during surveys and did not improve until shorelines became icebound and inaccessible. In future, the analysis of multiple mosaics for a single location should be considered to report on variability between sonar derived site-specific population estimates. Because these surveys are the only data available to describe the 2022/23 winter aggregations and the only data collected since 2018, analysis proceeded despite the known limitations.

Underwater camera survey

Andrews *et al.* (2020) conducted an underwater camera survey of SNS and bottom structure at the location of the main winter aggregation on Nov 27, 2019, the year after side-scan sonar data was collected on Dec 11, 2018. During that survey an underwater camera was affixed with underwater lights and towed near the river bottom for 2,681 m recording 109 min of video. During that survey 212 sturgeon were captured on camera of which every individual that could be clearly identified was confirmed as SNS. Since no other fish or sturgeon species were recorded, all side-scan sonar returns that had been recorded by Andrews *et al.* (2020) over the core aggregation in 2018 were counted as SNS. Following these results confirming single species occurrence, the underwater camera survey was not repeated for the main aggregation during this study.

The winter aggregation in Kennebecasis Bay was surveyed on 27 November 2022 during this study using an underwater camera to determine species composition. For this survey, a commercially available Vexillar® Scout camera with a DVR recording to a 32 GB microSD card was used with lights supplemented through the attachment of a scuba diving flashlight (bigblue $AL1200_{NP,}$ 1200 Lm, 10° beam). Each fish that appeared on camera was identified to species. The resulting ratio of SNS to other species was used as a correction factor for the side-scan sonar estimate. The bottom substrate was also described to make note of substrate type and character across the survey location.

Tagging and Winter Habitat Residency

To estimate percentages of the total SJR SNS population (fish ≥ 40 cm FL) aggregating in each identified winter habitat, we categorised the winter residency patterns of 18 SNS tagged with V16-4L 69 kHz acoustic transmitters (Innovasea Systems Inc.) in 2015 and described by Andrews *et al.* (2020). The tags used in the original study had an expected battery life of ten years and thus allowed those same tagged SNS to be continuously monitored in the SJR until this re-assessment (2022). This present study tabulated eight winters of data (2015/16-2022/23; 144 observations of winter location) for those individuals to calculate winter residency proportions in the SJR.

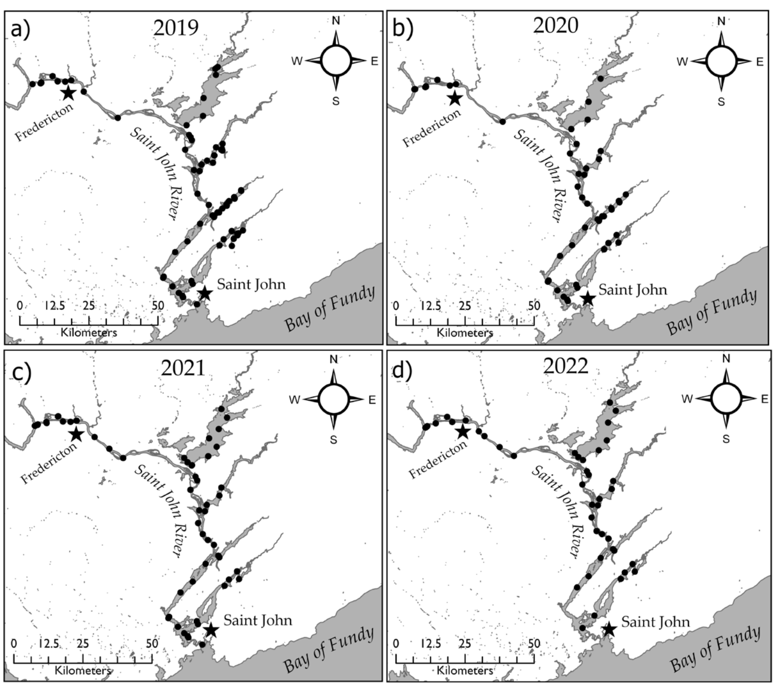

Fig 4 Annual positions of project specific VR2W acoustic monitoring stations positioned in the Saint John River including 93 in 2019, 53 in 2020, 85 in 2021, 48 in 2022 that could detect the movements of Shortnose Sturgeon *(Acipenser brevirostrum)* and resolve the winter habitat residency proportions between occupied winter refuges.

The tagging methodology in 2015 was as follows. Adult SNS (n=18, Total Length [TL] range = 100.5-128 cm) were captured by gill net in the SJR from 16-30 May 2015 (n=16 in Long Reach, n=2 in middle Kennebecasis Bay) and surgically implanted with V16-4L 69 kHz acoustic tags using an anesthetic of 40 mg/L solution of 10 parts ETOH: 1 part clove oil (eugenol). The sex of the tagged fish was not recorded. Tagged individuals were then tracked by a project-specific array of Innovasea® VR2W receivers (n=125 in 2015, n= 128 in 2016, n=135 in 2017 and n=60 in 2018; Andrews *et al.* 2020). Following this initial study period, the project specific receiver array numbered n=93 in 2019, n=53 in 2020, n=85 in 2021, and n=48 in 2022 (Fig 4) that were collectively able to identify annual winter habitat residency of all tagged individuals. The two SNS tagged in Kennebecasis Bay (~5 km from the overwintering location) were rarely observed to winter in the Kennebecasis. One individual occupied the Kennebecasis in two of eight years, the other was never observed in the Kennebecasis over winter.

Following eight years of continuous tracking, the mean proportional residency in each identified winter habitat was compiled. These relative proportions allowed for a projection of population abundance of SNS occupying un-surveyed habitats and to verify the accuracy of our abundance estimates when both prior estimates and side-scan derived enumeration became available for single locations (*i.e.* Kennebecasis). The number of SNS in each identified winter habitat within the SJR was calculated from these residency proportions as a mean percentage of the estimated total. When combined, these winter residency patterns allowed for a general estimation of the whole river population.

RESULTS

Side-scan Sonar

The survey of the main aggregation covered 342,392 m^2 where depths ranged from 4-9 m (mean = 6.5 m). Andrews *et al.* (2020) enumerated 12,284 SNS in 2018, with a kappa coefficient (k) = 0.98. Comparatively, 12,005 SNS polygons within the selection range of 40-150 cm were directly enumerated in the main aggregation on 24/25 November 2022. Our classification model produced a k = 0.975; almost identical to the 2018 assessment. These values for k demonstrate

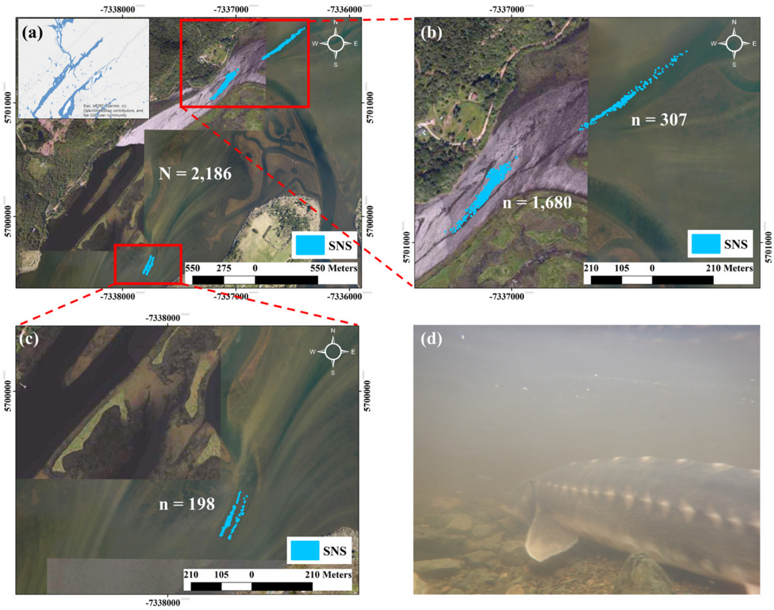

Fig 5 **An overview of the Kennebecasis study area along with the spatial distribution of the overwintering Shortnose Sturgeon (*Acipenser brevirostrum*)-SNS highlighted in blue (a). A fine-scale view of the core Kennebecasis aggregations is provided in (b) along with the number of SNS in each group. Similarly, a fine-scale view of the secondary channel grouping is shown in (c) along with an underwater image of a SNS in (d).**

strong agreement between the manually defined potential Shortnose Sturgeon and those selected by the sMLC.

Survey of the Kennebecasis aggregation covered an area of 85,510 m^2 (1/4 of the habitat extent of the main aggregation) where depths ranged from 2.5-7 m (mean = 4.5 m). Evaluation of side-scan sonar images resulted in the enumeration of 2,186 SNS polygons with the selection range of 40-150 cm that were spatially delimited within three distinct groups (see Fig 5a). Our supervised maximum likelihood classification (sMLC) rendered a $k = 0.979$, again nearly identical to that for the main aggregation in this study. The largest of the three defined groups contained 1,680 SNS polygons, the second largest group was located due northeast (directly upstream) of the largest group with 307 SNS polygons (Fig 5b). A third group was also present, located ~ 1.5 km (straight line measurement) due southwest from the other two groups in an adjacent side channel and contained 198 SNS polygons (Fig 5c). Of note is the river morphology characterizing the areas that aggregations occupied; each being denoted by the thalweg of a low gradient river.

Camera Survey

In total, the underwater camera survey in the Kennebecasis logged 58 minutes of video during which 229 sturgeon and two White Sucker (*Catostomus commersonii* Lacépède, 1803) were counted. All sturgeon detected in the camera survey were identified as SNS. Distance covered by the camera was not logged as the core winter aggregation in the Kennebecasis was confined to just 2,800 m^2 (0.6 sturgeon/m^2) and due to the lightweight camera, high current velocity, and low visibility, several passive drifts through the aggregation at different angles were repeated to collect the necessary footage. Surface water temperature at the time of the survey was 0.8°C.

Camera footage matched the side-scan sonar recordings which suggested very tight aggregation of SNS in this survey location (Fig 5). During several passes with the camera, the aggregation was observed to be so tightly associated that the camera became frequently

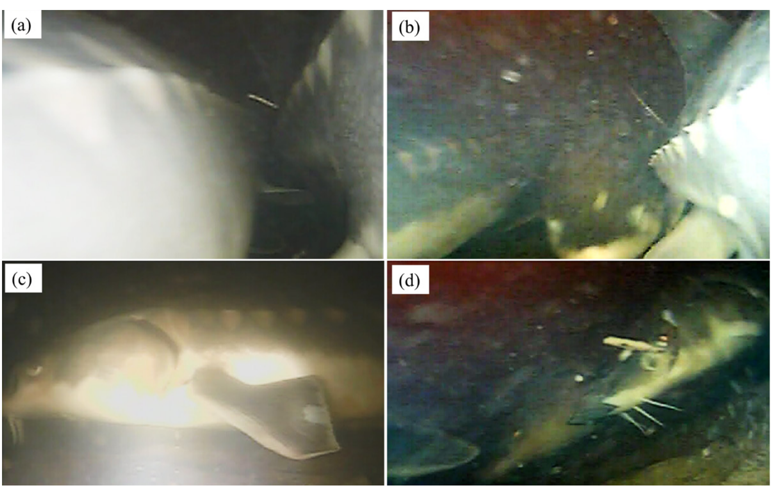

Fig 6 Still images extracted from underwater camera footage of the Shortnose Sturgeon (*Acipenser brevirostrum*) winter aggregation in the upper Kennebecasis Bay, Saint John River, New Brunswick taken on a Vexilar® Scout camera. Images provide an indication of the density of the aggregation and the stationary nature of the sturgeon under winter conditions. Panels include (a) camera stuck between two sturgeon, (b) camera caught between two sturgeon and blocked by a third, (c) sturgeon in profile in camera light, and (d) three sturgeon in profile showing close aggregation.

entrapped between fish (Fig 6a, b, d). Camera footage provided further evidence for the nearly immobile state of the SNS during the side-scan survey period. Observed SNS lay motionless on the bottom, no fin movement was observed, and they were undisturbed by the attached LED light (Fig 6c) or even physical contact from the camera itself. In several instances when the camera was inadvertently swept into the side of a sturgeon, no movement was observed, and sturgeon would eventually be either pushed sideways by the camera or the camera would need to be lifted over the fish to continue the survey. These observations provide further evidence that limited movement of SNS occurs during the side-scan survey transects described in this study. The substrate where SNS were observed in the Kennebecasis was clean sand; some rocks and plant debris were observed on bottom but only in areas where there was no sturgeon. No small/juvenile sturgeon appearing < 50 cm Fork Length (FL) were observed during this survey like observations made by Usvyatsov *et al.* (2012) in the same location.

Tracking and Population Estimate

Tagged SNS (n=18) were monitored by acoustic receivers over eight consecutive winters from 2015/16-2022/23 allowing for winter habitat residency proportions to be determined. In total, seven discrete overwintering locations were identified, increasing the number reported by Andrews *et al.* (2020) by two. These winter habitats included the main aggregation and the Kennebecasis aggregation, in addition to Grand Lake, Oromocto, Swan Creek, Washademoak Lake and an aggregation near Fredericton among those occupied by SNS (Table 1).

Following eight years of tracking, 144 winter positions were recorded for the 18 tagged SNS providing annual winter residency proportions within years for the duration of the tracking period (Table 1, 2). The eight-year mean proportional residency for the main aggregation was 64.6% while mean residency of the Kennebecasis was 9.7%. Occupancy of winter habitat near Fredericton also remained high as originally noted by Andrews *et al.* (2020) with an estimated 18.8% of Shortnose occupying that location. Over the eight-year monitoring period, nine of the tagged individuals (50%) were observed to switch from the winter habitat initially occupied in 2015. Four individuals (22%) occupied a different winter habitat from their main location of winter residency in one of the seven years of monitoring. Two sturgeon (11%) occupied a different location from their main

Table 1 Winter locations of tagged Shortnose Sturgeon (*Acipenser brevirostrum*) in the Saint John River, New Brunswick, monitored over eight consecutive years spanning winter 2015-16 to 2022-23. Tag numbers match tag IDs of Shortnose Sturgeon tagged by the Canadian Rivers institute in 2015 in ascending order. Total length (TL; cm) is the length of each Shortnose Sturgeon at the time of tagging in 2015. Observed winter locations include the main aggregation (MA; grey boxes), the City of Fredericton (Fred; blue boxes), Kennebecasis (Ken; yellow boxes), Washademoak Lake (Wash; green boxes), the vicinity of Swan Creek (~Swan; pink boxes), a single observation in Grand Lake (Grand L; red box), and a single observation near Oromocto (orange box).

		Winter							
Tag	TL (cm)	2015-16	2016-17	2017-18	2018-19	2019-20	2020-21	2021-22	2022-23
1	102.1	MA	MA	MA	MA	MA	MA	MA	MA
2	102.9	Ken	MA	MA	MA	MA	MA	MA	MA
3	106.5	MA	Ken	MA	Ken	MA	MA	MA	MA
4	100.5	MA	Wash	MA	MA	MA	MA	MA	MA
5	128.5	MA	MA	MA	MA	MA	MA	MA	MA
6	116	MA	MA	MA	MA	MA	Grand L	MA	MA
7	106.6	MA	MA	MA	MA	MA	MA	MA	MA
8	104.1	Ken	MA	MA	MA	MA	MA	MA	MA
9	119.4	MA	Fred	Fred	Fred	Fred	Fred	MA	MA
10	108	MA	MA	MA	MA	MA	MA	MA	MA
11	109.2	MA	MA	MA	MA	MA	MA	MA	MA
12	116.8	Ken	Ken	Ken	Ken	Ken	Ken	Ken	Ken
13	106.7	MA	MA	MA	MA	MA	MA	MA	MA
14	106.7	MA	Wash	MA	Wash	MA	Wash	Wash	Wash
15	121.9	Fred	Fred	Fred	Fred	Fred	Fred	Fred	Fred
16	105.4	~Swan	~Swan	Ken	MA	MA	MA	Ken	Oromocto
17	109.2	Fred	Fred	Fred	Fred	Fred	Fred	Fred	Fred
18	111.8	Fred	MA	Fred	MA	Fred	Fred	Fred	Fred

location of winter residency in two of the eight years of monitoring. The remaining three individuals (17%) made more frequent switches between winter habitats across monitoring years (Table 1). Only one tagged sturgeon switched between more than two key winter locations. Habitat switching only occurred between winter periods and was never observed during winter.

Due to habitat switching, residency proportions in the main aggregation ranged from 55.6% in 2016 to 72.2% in 2019 (mean 64.6%) across eight years of monitoring (Table 2). At this site, the side-imaging sonar-based enumeration counted 12,005 individual SNS. Proportional residency at the Kennebecasis aggregation ranged from 5.6% observed in three of the monitoring years to 16.7% in 2015 (mean 9.7%) across eight years of monitoring (Table 2). In this location, side-imaging sonar provided an enumeration of 2,186 individuals across three spatially discrete aggregations. In combination, these two surveyed locations represent on average 74.3% of the SNS population occupying the SJR across years resulting in an estimated total adult population of 19,100 SNS (Table 2).

Table 2 **Winter residency observations of tagged Shortnose Sturgeon (*Acipenser brevirostrum*) monitored from winter 2015-16 to 2022-23 indicating the proportion of tagged sturgeon occupying each identified winter habitat location within and across years. The number of Shortnose Sturgeon in the main aggregation and Kennebecasis aggregations indicated by the * represent the enumeration from side-scan sonar surveys conducted in this study. In combination, these two aggregations which contained 14,191 sturgeon represent an estimated 74.3% of the total. This ratio was used to calculate the whole river (total) population of 19,100 from which estimates of the five other aggregations (Fredericton, Oromocto, Washademoak, Swan Creek, and Grand Lake) were back calculated. The back calculation was conducted using the average percentage of tagged Shortnose Sturgeon (in winter habitats) multiplied by the total.**

Overwintering Location	2015	2016	2017	2018	2019	2020	2021	2022	Average percentage of tagged Shortnose Sturgeon	Mean local population calculated from average percentage or enumeration (*)
Main Aggregation	61.1%	55.6%	66.7%	66.7%	72.2%	61.1%	66.7%	66.7%	64.6%	12,005*
Kennebecasis	16.7%	11.1%	11.1%	11.1%	5.6%	5.6%	11.1%	5.6%	9.7%	2,186*
Fredericton	16.7%	16.7%	22.2%	16.7%	22.2%	22.2%	16.7%	16.7%	18.8%	3,582
Oromocto	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	5.6%	0.7%	133
Washademoak	0.0%	11.1%	0.0%	5.6%	0.0%	5.6%	5.6%	5.6%	4.2%	797
Swan Creek	5.6%	5.6%	0.0%	0.0%	0.0%	0.0%	0.0%	0.0%	1.4%	265
Grand Lake	0.0%	0.0%	0.0%	0.0%	0.0%	5.6%	0.0%	0.0%	0.7%	133
Total Population	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**100%**	**19,100**

From this total estimate, a back calculation of the proportional occupancy for each other winter habitat yields 3,582 SNS wintering near Fredericton, 797 in Washademoak, 265 in the vicinity of Swan Creek, and 133 in both Oromocto and Grand Lake (Table 2). Two White Suckers were observed among the Kennebecasis SNS aggregation, their small size < ~30-40 cm FL would have likely rendered them undetectable by the sonar at the survey depth. Fish of that length were further removed by the classification model and our estimates were therefore not corrected by the calculated 0.87% ratio (2 White Sucker among 231 observed fish of which 229 were SNS).

Using only the observed winter proportions from the study year of 66.7% of SNS in the main aggregation and 5.6% in the Kennebecasis (Table 2), the estimated total would be 19,628. While the 2018 estimate total was reported as 20,101 SNS >~40 cm FL. The lower total calculated during this study could be the result of error during image collection, processing, and filtration. Although winter residency proportions produced from a small group of 18 tagged sturgeon can also affect the calculation. During tracking efforts, no more than three and most often only two of the tagged SNS occurred in the Kennebecasis in any sampling year resulting in variability in the proportional occupancy of that location (ranging 5.6-16.7%). If a larger number of tagged SNS had been available for assessment these residency proportions could have been more refined.

DISCUSSION

Side-scan sonar has most commonly been used to map and assess benthic habitat (Switzer *et al.* 2020), although previously studies have already employed this technology to map, count, and even measure sturgeon (Flowers & Hightower 2013, Kazyak *et al.* 2020). When sonar is applied to create a population estimate of sturgeon such as SNS over a broad area, two significant challenges arise. First, SNS are active during the warmer months of the year (Dadswell 1979) and second, large areas cannot be efficiently surveyed without inevitably and inadvertently double counting some individuals and missing many others. These challenges are avoided by conducting surveys in winter when SNS are tightly aggregated and in a state of torpor. Winter is a particularly effective time for side-scan sonar surveys since SNS appear to maintain close bottom association, therefore sonar derived acoustic shadows are eliminated and the resulting sonar returns are easier to identify, count, and measure.

The enumeration produced in this study for the main SNS aggregation in SJR (n=12,005) is only 2% (n=279) different than the final count of 12,284 made in the same location in 2018. In consideration of the low occurrence of winter habitat switching between years (Table 1) and the omission of individuals within the enumeration process due to sonar pass overlap and length-based data filtering, these numbers appear to at least suggest a stable abundance within the annual aggregation. The 2018 side-scan enumeration of the main winter aggregation and proportion-based population estimate resulted in a predicted Kennebecasis aggregation size of 2,513 individuals (Andrews *et al.* 2020). Our 2022/2023 survey of the Kennebecasis aggregation (this study) enumerated 2,186, approximately 300 individuals different than what was estimated four years prior. Although we suspect that the count produced in the 2022/23 survey was an underestimate due to filtering of individuals following sonar data stretching, possible inaccuracies from counting tightly packed SNS, and the likely occurrence of other small aggregations in side-channels that were not located during this survey. Local fishing guides commonly reported one to two SNS aggregations in late autumn separate from the core Kennebecasis aggregation (S. Delaney, Saint John River fishing guide, pers comm). This phenomenon has previously been observed by the authors and was recorded in this study.

Following the 2018 survey and without direct sonar enumeration, Andrews *et al.* (2020) suggested that a sonar and acoustic telemetry-based estimate of 2,513 SNS occupying the Kennebecasis were comparable to the estimates of 4,836, 3,852 and 5,222 reported from winter video surveys in 2005, 2009, and 2011 (Li *et al.* 2007, Usvyatsov *et al.* 2012). Now having directly enumerated the Kennebecasis aggregation as n=2,186 individuals it is more likely that those prior studies reported overestimates, particularly because 198 (9%) of the SNS reported in this study were not associated with the core aggregation, and therefore would have been missed in 2005, 2009 and 2011. Since the video-based surveys described by Li *et al.* (2007) and Usvyatsov *et al.* (2012) were conducted over the span of months, sturgeon would have been able to redistribute themselves as they are observed to do during each tidal cycle, inevitably leading to repeated counts. Although, this study has shown that winter residency proportions can shift between assessment years.

The acoustic tracking component of this study estimates that the Kennebecasis supports only ~10% of the overall SNS population (>~40 cm FL) between years. Due to this low proportion and limited winter habitat switching, the recreational catch and release fishery conducted in this region from September to late-November and briefly at ice out in early March may have limited ability to impact on the overall SJR SNS population. Although stress related responses during an annual one-day angling derby are reported (Struthers *et al.* 2018). However, if the earlier population estimates (Li *et al.* 2007, Usvyatsov *et al.* 2012) and the current estimate (this study) are in fact accurate, unbiased, estimates of the SNS population at the Kennebecasis site independent of residency proportions, a potentially concerning alternative finding might be that the popular catch-and-release fishery in the Kennebecasis is either reducing sturgeon numbers through mortality or causing departure from this aggregation following disturbance. Only carefully conducted telemetry studies examining the effects of repeated capture in the recreational fishery could rule out the possible fisheries related impacts.

Across seasons, 3-4 of the tagged SNS (16.7-22.2 %) consistently wintered near Fredericton. Without a dedicated survey of the Fredericton location, it is not possible to determine if these individuals are a true indicator of a large aggregation near the city, or rather if the small sample size resulted in a few individuals being tagged that happened to occupy that location annually. If these estimates are accurate, the main aggregation, Kennebecasis, and Fredericton aggregations would total 93.1% of the adult population and in combination, could provide a very robust combination of survey sites.

During the 2019 survey of the main winter aggregation (Andrews *et al.* 2020), a length distribution curve was created for the enumerated SNS. These data approximated findings of Dadswell (1979), but these original measurements were collected after the completion of Mactaquac Dam in 1968 and may not have reflected the length frequency distribution of an unimpacted SNS population. Dadswell (1979) also described SNS reaching maturity in the SJR from 12-18 years at 50 cm FL (Bain 1997, Bain et al 2007), while Usvyatsov *et al.* 2012 documented the smallest SNS in the Kennebecasis winter aggregation as 54 cm FL. Following these findings and the suspected limitations of our applied sonar method in resolving accurate length (Andrews *et al.* 2020), it is likely that the identified winter habitats contain the known adult population of SNS in the SJR along with a small number of older but immature fish. No data exists on where juvenile SNS < 40 cm FL might occur in the SJR and their abundance is not included in our estimate.

The small sample size of monitored individuals resulted in shifting winter residency proportions across occupied habitats, but we suspect that larger more stable winter habitats consistently retain larger aggregations than smaller, less stable locations. Therefore, regardless of overall abundance, each winter habitat should support a consistent proportion of the overall adult population even if the overall population should vary. Monitoring of one or two large and easily surveyed locations may be sufficient to inform upon the status of the entire population even when estimating total abundance is not possible. Although, more detailed work will be required to determine if population demographics are changing despite an apparent stable abundance. Among the surveyed SNS, some individuals were never observed to switch between winter habitats while others switched frequently. No pattern in relation to spawning and non-spawning years was discernable. If more SNS are tagged in the future these data may refine estimates of proportional winter residency in the SJR.

LIMITATIONS

This study was subject to several limitations that should be considered and improved upon should these monitoring efforts continue in the future. Due to this small sample size of telemetered SNS, shifts of one or two individuals in winter could result in large changes in residency proportions and changes to the ensuing population estimate. This could be fixed by including a larger sample size of acoustically tagged SNS in the future, but considering the data at hand, greater confidence should be placed in the sonar enumerations than the projected proportions until a more robust tracking dataset can be included for analysis. Since the sonar transects were driven by hand, the boat speed, direction, and overlap of the side-scan sonar coverage between transects could not be precisely maintained which occasionally led to image distortion and stretching and could have affected the image filtration process. Movement of SNS could also have resulted in missed individuals or duplicate counts, but we know that movement by SNS is minimal within tidal cycles (Fig 6). As such, we must re-iterate the importance of completing passes during a single outgoing tide and at the coldest possible water temperature to ensure that sturgeon movement is kept to an absolute minimum during the survey.

Without multiple clear side imaging mosaics of each location, this study was unable to produce an estimate of variance around the reported population estimate. Three scans were collected for both the main aggregation and the Kennebecasis aggregation, but windy conditions resulted in only the single clearest scan being analysed. In the future, provisions should be made to gather multiple data sets to accurately describe error associated to the population estimate when weather conditions permit. Limitation are also apparent in the ability to resolve highly accurate length data, and while SNS abundance appears to have remained stable, further analysis must be made to determine if population demographics are changing over time. Finally, the accuracy of the sMLC classification could have been biased by the hand selection of "sturgeon" and "non-sturgeon" polygons that may have categorized the best examples of representative pixels from each group. Randomly selecting targets to delineate while training the sMLC might reduce bias in the selection and subsequent classification process.

CONCLUSIONS

Side-scan sonar enumeration of SNS is a rapid and effective method to quantify SNS occupying winter habitats. The estimate produced for the main aggregation during this study in winter 2022/23 was only 2% different than the number calculated for the same location in 2018. Predictions of abundance in the Kennebecasis in 2018 were only 327 fish greater than the count generated for that aggregation within this study. This difference may be accounted for by the potential removal of individuals within the automated data filtration process following sonar data distortion or shifting residency proportions. Despite such limitations, the rapid enumeration of SNS in winter aggregations by commercially available side-scan sonar appears to be a method which could be refined to produce a reliable and repeatable population estimates for SNS in the SJR in the future. Continued use and development of this method may therefore provide a viable solution to long-term monitoring to support management under the Fisheries and Oceans Canada (2016) management plan. Following these results and those of Andrews *et al.* (2020), there is growing evidence that the abundance of adult SNS in the SJR has likely remained stable since the initial surveys by Dadswell (1979) but more work is needed to determine if population demographics have also remained stable.

Acknowledgements We would like to thank Fisheries and Ocean Canada for funding the second iteration of this study and Jeremy Broome for providing project oversight. We thank Elizabeth Bateman for assisting with side-scan sonar mapping, underwater camera surveys, and survey organization. We would also like to thank Daniel Arluison for tagging the 18 Shortnose Sturgeon tracked in this study as part of the Mactaquac Aquatic Ecosystem Study (MAES) and the technicians at the Canadian Rivers Institute (CRI) at the University of New Brunswick, Fredericton, Canada for maintaining the Saint John River receiver array over the past eight years. The MAES program is largely funded by the New Brunswick Power Corporation and NSERC. We would also like to thank Steve Delaney for the generosity with which he has provided his hard-earned knowledge on the habitat and habits of Shortnose Sturgeon in the Kennebecasis and for his loan of the underwater camera that made this work possible.

Author contributions: S.N. Andrews planned the project and coordinated the boat and equipment required for data collection. S.N.A drove the side-scan transects of both the main Shortnose Sturgeon aggregation and the Kennebecasis aggregation and conducted the underwater camera surveys of the Kennebecasis aggregation. S.N.A compiled the side scan mosaics for export to GIS, analysed the video footage, and analysed an 8-year acoustic tracking data set to determine the proportional habitat occupancy and mixing rates between aggregations. A.M. O'Sullivan conducted the remote sensing statistical analysis to enumerate Shortnose Sturgeon in each surveyed aggregation and produced model derived Figs. R.A.C and T.L oversaw and acquired funding for the original tagging and monitoring effort in 2015 and have continue to oversee the acoustic river monitoring, receiver placement, and database management for the incurred detections.

S.N.A and A.M.O.S wrote the study results that were reviewed by all authors.

Conflict of Interest Statement: The authors declare there are no competing interests.

Data Availability Statement: To protect identified Shortnose Sturgeon winter aggregations from exploitation no shared data is available for this study.

Funding: This project including work by S.N. Andrews and A.M. O'Sullivan was funded by Fisheries and Ocean Canada, Maritimes Region, Species at Risk Program. The maintenance of the Saint John River Acoustic receiver array and the original tagging of the Shortnose Sturgeon tracking in this study was funded in part by the New Brunswick Power corporation with matched funding from NSERC-CRDPJ 462708-13.

Impact Statement: The use of side-scan sonar to enumerate winter aggregations of Shortnose Sturgeon provides a rapid, repeatable, and low-cost option for assessing this species of concern.

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