Impacts of marine shipping on the underwater soundscape and Chinook salmon behaviour

by

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B.Sc., University of Wisconsin-Superior, United States, 2012

M.Sc., University of Minnesota, United States, 2016

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We acknowledge and respect the Lək̓ʷəŋən (Songhees and Esquimalt) Peoples on whose territory the university stands, and the Lək̓ʷəŋən and W̱SÁNEĆ Peoples whose historical relationships with the land continue to this day.

Supervisory Committee

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Abstract

With the persistent growth of the shipping industry, the underwater soundscape across the globe has been changing. The prevalence of low-frequency noise from shipping activities has led to a noticeable increase in ambient underwater noise levels. Research to date has focused on large commercial vessels in motion, but commercial vessels are also anchoring in coastal ecosystems and there is a high presence of small commercial vessels that might be altering the marine soundscape. In this dissertation, these underrepresented anthropogenic noise sources are explored in Cowichan Bay, and Campbell River, British Columbia using underwater hydrophones. Results from these chapters (2 and 3) demonstrated significant changes to the underwater soundscape from anchored commercial bulk carriers and tugboats, highlighting the need to understand these noise sources further. Additionally, the growing body of literature on the impacts of anthropogenic noise on the underwater soundscape has generated concern about the impacts of the elevated noise on marine species. In the Northeast Pacific Ocean, many ecologically important species have been declining since the 1970s including Chinook salmon (*Oncorhynchus tshawytscha*). Although behavioural changes including modified sound production behaviours have been documented in response to anthropogenic activities for some fishes, there has been little research on the impacts of shipping on Chinook salmon. Furthermore, Chinook salmon sound production has received little attention and only limited information on frequency and amplitude has been reported. To better understand the impacts of shipping on Chinook salmon vocal behaviour, understanding of salmon sound production, Chinook and other Pacific salmon, is required. Sound production in three species of Pacific salmon (Chinook, pink, *O. gorbuscha* and coho salmon, *O. kisutch*) was evaluated at Big Qualicum Hatchery (chapter 4). Chinook and coho salmon were found to produce air movement, hydrodynamic and pulse sounds, while pink

salmon were shown to likely produce all these sounds. However, because pink salmon were always recorded in mixed schools with Chinook salmon, further work is required to validate their sound production. Additionally, the impacts of shipping on Chinook salmon movement and behaviour have never been evaluated, but vessel noise and other anthropogenic sources like pile driving have been shown to produce spatial displacement and behavioural changes in other salmonids. To begin to address this issue, changes in Chinook salmon behaviour in the presence of shipping noise were evaluated using acoustic tags in Cowichan Bay, British Columbia (chapter 5). Significant changes in depth for tagged salmon were observed with increased sound pressure levels and number of AIS-equipped vessels present. However, the influence of the number of AIS-equipped vessels present on the overall activity levels of Chinook did not appear linear and further work is needed to understand vessel noise effects on activity levels. This chapter represents the first study demonstrating the impacts of increased underwater noise levels on Chinook salmon. Collectively this dissertation highlights the impacts of anchored commercial vessels and tugboats on the underwater soundscape and the influence of shipping noise on Chinook salmon behaviour, demonstrating the importance of developing mitigation methods to help reduce the noise produced by shipping activities to protect these ecologically, and culturally important species.

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Dedication

To my big sister, Lyndsie.

Thank you for showing me the beauty in the world and how to appreciate the small things.

Love you forever!

Chapter 1 - Introduction

The underwater soundscape is an important component of marine ecosystems, and is composed of sounds produced by geological, biological, and anthropogenic sources (Pijanowski et al., 2011). Geological sounds are various types of "natural" sounds that occur in a habitat or at specific locations, such as wind, rain, and surf, while biological sounds are natural sounds that are produced by animals such as invertebrates, fish, and mammals. In contrast, anthropogenic sounds are from human-made sources, including sounds from boats, seismic surveys and pile driving (reviewed in Duarte et al., 2021). Water is an efficient conductor of sound; sounds travel faster and longer distances in water compared to in air (Au and Hastings, 2008). For example, low frequency sounds (57 Hz) projected at Heard Island in the Southern Indian Ocean were recorded to travel around the globe, with the sound detected off both the west and east coasts of North America (Munk et al., 1994). Aquatic animals use this characteristic to their advantage by producing sounds for communication (Fine, 1978; Ford, 1989) as well as to find and capture prey (Au et al., 2004).

All marine mammal species are known to produce sounds and their sound production has been extensively studied worldwide (May-Collado et al., 2007). Bottlenose dolphins (*Tursiops truncatus*) produce signature whistles which they use to communicate and identify each other (Janik et al., 2006; Janik and Slater, 1998). Similarly killer whales (*Orcinus orca*) produce a variety of social calls (Ford, 1989). Male humpback whales (*Megaptera novaeangliae*) produce vocalizations used to attract females (Herman et al., 2013) and male harbour seals (*Phoca vitulina*) produce a 'roar' sound during the mating season (Nikolich et al., 2016). In addition to sounds used for communication, some marine mammals also produce sounds for prey localization (Au et al., 2004). Killer whales produce broadband high frequency (20 - 80 kHz) click trains, which are used to locate prey and perceive their surroundings (Au et al., 2004). Other odontocetes produce similar sounds, harbour porpoise (*Phocoena phocoena*) have broadband click trains with peak frequencies between 125 and 130 kHz (Au et al., 1999) and false killer whales (*Pseudorca crassidens*) have peak frequencies between 88 -104 kHz (Au et al., 1995).

Fish have also been documented to produce sounds, with 1,185 different species across 133 families recorded to date (Looby et al., 2023, 2022). Atlantic cod (*Gadus morhua*) produce low frequency vocalizations during spawning and agonistic behaviours (Hernandez et al., 2013). Similarly, oyster toadfish (*Opsanus tau*) produce two distinct vocalizations, boat-whistle and grunt (Fine, 1978; Maruska and Mensinger, 2009; Tavolga, 1958). In addition to communication sounds, some fish species produce incidental sounds. For example, Pacific herring (*Clupea pallasii*) and Pacific sardine (*Sardinops sagax*) produce sounds with frequency ranges of 320-800 Hz and 380-850 Hz, respectively, and have peak production at midnight (Kuznetsov, 2009), but what these sounds are used for is unclear.

Sound production has also been observed in multiple invertebrate species (Coquereau et al., 2016). Snapping shrimp (*Synalpheus paraneomeris*) produce loud clicks (up to 190 dB re 1 μPa) from quickly closing their frontal claws, these clicks are typically between 2 and 5 kHz but can be up to 200 kHz (Au and Banks, 1998). New Zealand paddle crab (*Ovalipes catharus*) have been observed to produce three sounds, a rasp (peak frequency: 4,876-10,000 Hz), zip (peak frequency: 662 Hz) and bass (peak frequency: 45 Hz) and the zip and bass were associated with courtship behaviour (Flood et al., 2019). Additionally, great scallops (*Pecten maximus*) produce impulsive sounds (3-48 kHz) when contracting their adductor muscles, and closing their valves (Di Iorio et al., 2012).

The underwater soundscape has been changing globally due to anthropogenic noise (Hildebrand, 2009). Noise levels (< 1000 Hz) in the Northeastern Pacific Ocean have been increasing by ~3 dB per decade for the last four decades up to early 2000s (McDonald et al., 2006). One main contributor to this increase is the shipping industry, which has increased four-fold between 1992 and 2012 (Tournadre, 2014), and is predicted to continue growing until 2050 (Sardain et al., 2019). Large commercial vessels have been documented to account for 30% to 90% proportion of the underwater soundscape depending on the time of year, with highest contribution in the summer months (Burnham et al., 2021). Additionally, the COVID-19 global shutdown demonstrated the impact of anthropogenic activities on marine ecosystems when these activities were absent (Bates et al., 2021; Pine et al., 2021; Robinson et al., 2023). For example, there was a reduction in trade into and out of Canada, which caused a reduction in the number of commercial vessels and subsequently reduced underwater noise levels (Thomson and Barclay, 2020).

1.1 Vessel noise

Sources of vessel noise can be broken down into two major categories, commercial (*i.e.*, non-pleasure use, paying customers or paid crew) and recreational vessels (*i.e.*, pleasure use or daily living). Large commercial vessels vary in the source levels produced with container ships and bulk carriers producing the loudest at mean source levels (20-1000 Hz) of between 184.2 dB re 1 μPa^2 to 188.1 dB re 1 μPa^2 respectively, while oil and product tankers produce are lower at source levels between 178.5 and 181.3 dB re 1 μPa^2 respectively (McKenna et al., 2012). The source level of these vessel can increase sound levels (<15 kHz) by as much as 20 dB above ambient levels (Veirs and Veirs, 2006). For vessels in motion, most of the noise is from propeller cavitation, generating tonal harmonics and broadband noise due to blade rate and cavitation bubble collapse (Wittekind and Schuster, 2016). Propeller cavitation is the primary cause for low frequency noise (< 300 Hz), and peaks at around 50 Hz (Wittekind and Schuster, 2016). However, generators and other machinery also produce noise that is detected underwater (Arveson and Vendittis, 2000). Noise at the frequencies of 24 and 30 Hz has been linked to diesel generators, and power lines have been observed to produce noise around 60 Hz (Arverson and Vendittis, 2000).

The sound levels produced by commercial vessels are also influenced by their speed, with vessels moving faster producing more noise compared to slower moving vessels (Arveson and Vendittis, 2000; McKenna et al., 2013; Ross, 1976). For example, a bulk cargo ship traveling at 8 knots had a source level of 178 dB re 1 µPa @ 1 m compared to 192 re 1 µPa @ 1 m when it was traveling at 16 knots (Arveson and Vendittis, 2000). When modeling source level of container ships, vessel speed was the best predictor (McKenna et al., 2013). Vessel speed also influences ambient sound pressure levels, with vessel speed increasing underwater sound levels by between 0.2 and 2.3 dB/knot (Halliday et al., 2020).

Recreational vessels are much more difficult to examine due to the lack of tracking (*e.g.*, Automatic Identification System, AIS) for each individual vessel, however, recently more research has been conducted to examine the noise produced by recreational boats and their impact to the soundscape (Picciulin et al., 2022; Wilson et al., 2022). Source levels (50 – 20,000 Hz) of recreational vessels are highly variable and range from 147 dB re 1µPa @ 1m up to 163 dB re 1µPa @ 1m for typical cruising speed (5 – 19 knots), but increased to source levels of up to 180 dB re 1µPa @ 1m for maximum speeds of up to 25 knots (Picciulin et al., 2022). In New Zealand, small recreational vessels were found to significantly increase the underwater soundscape for low frequencies (< 800 Hz), but varied by site and season (Wilson et al., 2022).

Similarly in the Canadian Arctic, small vessels increase sound pressure levels by 3.4 to 7.5 dB re 1µPa in various frequency bands between 50 and 24,000 Hz (Halliday et al., 2020).

In the waters around Vancouver Island, there is a high presence of ships throughout the year (Erbe et al., 2012) and on average 19.5 ships that pass through Haro Strait every day (Veirs et al., 2016). These high shipping areas create noise hotspots in Haro Strait, Juan de Fuca, and Johnstone Strait (Erbe et al., 2014), which overlap with critical habitat for ecologically important species, including resident killer whales (Ford et al. 2017) and Chinook salmon (*Oncorhynchus tshawytscha*). These increased noise levels have raised concern regarding the impacts these noise levels have on marine species that rely on sound for communication and foraging (Cox et al., 2018; Gomez et al., 2016; Murchy et al., 2020), and have led to increased research on the topic.

1.2 Impacts of vessel noise on marine species

The need to understand the impacts of vessel noise on marine animals has led to a growing body of literature on the subject with focus starting on marine mammals (Erbe et al., 2019). Beluga whales (*Delphinapterus leucas*) have longer dives and increase their speed in the presence of vessels (Blane and Jaakson, 1994), similarly humpback whales spend more time under water (longer dives) and showed horizontal avoidance in the presence of whale watching vessels (Stamation et al., 2010). However, some marine mammals (*e.g.,* bowhead whales, *Balaena mysticetus;* and North Atlantic right whales, *Eubalaena glacialis*) do not exhibit behavioural responses to vessels (Martin et al., 2023; Nowacek et al., 2003) which could lead to an increase in vessel strikes. In addition to movement behaviours, vessel noise has also been shown to change vocal behaviour in marine mammals, with an increase in noise levels increasing call amplitude and shifting frequencies in many species (Castellote et al., 2012; Dahlheim and Castellote, 2016; Holt et al., 2009).

Fish are also impacted by human-generated noise, which causes behavioural and vocal changes in many fish species (Holt and Johnston, 2014; Ivanova et al., 2020; Luczkovich et al., 2016; Mills et al., 2020; van der Knaap et al., 2022). Arctic cod (*Boreogadus saida*) have been shown to move away from vessels and limit exploratory behaviours when vessels are in close proximity (Ivanova et al., 2020). However, behavioural responses are species specific with some fish species hiding instead of moving away from vessel noise (Mills et al., 2020). Vessel noise has also been shown to alter feeding and antipredator behaviours in fish (Simpson et al., 2015; Vasconcelos et al., 2007; Voellmy et al., 2014b, 2014a). For example, three-spined stickleback (*Gasterosteus aculeatus*) made more unsuccessful strikes on *Daphnia* when exposed to noise, and European minnow (*Phoxinus phoxinus*) decreased feeding while exposed to the same noise source (Voellmy et al., 2014a). Mortality due to predation also increases during noise disturbances. European eels (*Anguilla anguilla*) were less likely to startle in response to a simulated predator when exposed to noise, and when they did startle, they were slower and caught more quickly compared to controls (Simpson et al., 2015). Similar to vocal changes observed in marine mammals, fish species also alter the amplitude and frequency of their vocalizations (Brown et al., 2021; Holt and Johnston, 2014; Luczkovich et al., 2016).

Invertebrates have been relatively understudied for effects of vessel noise on behaviours, but recent work has shown that invertebrates are impacted by vessels (Murchy et al., 2020). Spiny lobsters (*Palinurus elephas*) had higher velocity and distance moved when exposed to boat noise in tanks (Filiciotto et al., 2014), while common prawn (*Palaemon serratus*) spent more time outside of their shelters and resting during playback of boat noise (Filiciotto et al., 2016). Vessel noise has also been shown to decrease ventilation in Hummingbird bobtail squid

(*Euprymna berryi*; Putland et al., 2023), and increase visual displays (*e.g.*, change colour more often) in common cuttlefish (*Sepia officinalis*; Kunc et al., 2014).

Shipping noise has been observed to invoke a stress response in a variety of taxa (Celi et al., 2015; Nichols et al., 2015; Rolland et al., 2012). Marine mammals, including North Atlantic right whales, display a stress response due to shipping noise which has been documented using molecular techniques (Rolland et al., 2012). Various fish species have shown a stress response in the presence of noise created by ships (Wysocki et al., 2006). For example, gilthead sea bream (*Sparus aurata*) blood cortisol levels increased when exposed to vessel noise playback for 10 days (Celi et al., 2016). The intensity of cortisol elevation appears to be correlated with decibel level (Nichols et al., 2015). Similarly, marine invertebrates exhibit a stress response to shipping noise documented through measures of gene expression and haemolymph (Celi et al., 2015).

Population level impacts are another component of the impacts of underwater noise on marine species that has only been minimally explored (Hin et al., 2019; Pirotta et al., 2018; Soudijn et al., 2020). A Population Consequences of Disturbance (PCOD) framework can be used to take individual level impacts of underwater noise and relate them to population level consequences (Pirotta et al., 2018). Models using Atlantic cod (*Gadus morhua*) populations demonstrated that noise pollution reduces population growth because during noise exposure less food is consumed and foraging is altered (Soudijn et al., 2020). Population growth is reduced in marine mammals when exposed to noise (Hin et al., 2019), demonstrating population impacts across multiple trophic levels. Additionally, there has been little research on what additional impacts increased noise could have on predator-prey dynamics and how predators, like southern resident killer whales, are affected by noisier foraging environments.

1.3 Endangered resident killer whales and their prey, Chinook salmon

Southern resident killer whales (*Orcinus orca*) are a small population of fish-eating killer whales that live off the west coast of Canada and the United States (Ford et al., 1996). They are subdivided into three distinct pods, J pod, K pod and L pod (Ford, 1989). Southern resident killer whales range from Monterey Bay, California to Chatham Strait, Alaska (Ford et al., 2017). However, all three pods typically spend their summer in the Salish Sea (waters between Washington State, USA and British Columbia, Canada, including waters surrounding Puget Sound, the San Juan Islands, and Vancouver Island), with Haro Strait being a common region frequented by all pods (Hauser et al., 2007). Beginning in the 1960s and 1970s the population of southern resident killer whales has fluctuated, beginning with a removal of at least 56 individuals for marine parks and resulting in the death of approximately 10 more due to capture procedures (Asper and Cornell, 1977). Southern resident killer whales are currently listed as endangered on the Species at Risk Act (Canada) and the Endangered Species Act (United States) and have a population of approximately 73 individuals (Center for Whale Research, 2023).

Resident killer whales primarily feed on salmonids (96%), with Chinook salmon being the main salmonid consumed (Ford and Ellis, 2006). Chinook salmon are the largest salmonid species and have a high protein (~19.6%) and lipid (~10.5%) content, giving them a high caloric value for resident killer whales (O'Neill et al., 2014). However, lipid content appears to vary based on run timing and spawning location, with earlier runs (*e.g.,* Fraser River Spring 5_2) having higher lipid content compared to later runs (*e.g.,* Fraser River Fall 4_1; Lerner and Hunt, 2023). Resident killer whales consume mostly Chinook salmon in all months except for October and November when Chinook salmon are spawning (Ford et al., 2009).

However, Chinook salmon are the least abundant of the Pacific salmon species within the southern resident killer whales' range (Heard et al., 2007) and their populations have been declining (Riddell et al., 2018). In southern British Columbia, 13 Conservation Units have declined by more than 50% in spawner abundance in the last 12 years, 7 declined by over 70% and one has declined by over 90% (Riddell et al., 2013). Chinook salmon have also been declining in size-at-age since the end of the 1970s, with age four and five individuals declining by 7% and 9% respectively (Ohlberger et al., 2019). These declines are resulting in less, and smaller individuals returning to spawn and as available prey.

Southern resident killer whales and Chinook salmon show a typical predatory-prey interaction, with predators and prey cycling through time (Murdoch et al., 2003). In this case, Chinook salmon abundance shows a strong link to resident killer whale mortality, with increased mortality in the killer whale populations following one year behind low salmon numbers (Ford et al., 2009). Specific stocks and regions are also linked to body condition and mortality in southern residents, with higher abundance in Fraser River (Salish Sea region) and Puget Sound stocks decreasing the probability of body condition decline (Stewart et al., 2021). Additionally, salmon availability has a positive relationship with reproduction and fecundity in killer whales; in years with fewer Chinook salmon the probability of a female killer whale giving birth was 50% lower compared to high salmon abundance years (Ward et al., 2009).

Foraging theory predicts switching away from the 'optimal' prey source when prey abundance is low (Mittelbach and McGill, 2019), however, southern resident killer whales do not exhibit any evidence of prey switching. Southern residents continue to select for Chinook salmon, even when numbers are low demonstrating no shift in prey selection based on availability (Ford et al., 2009). However, the number of days southern residents are present in the

Salish Sea has been declining and is correlated to decreased Chinook salmon abundance in the area (Stewart et al., 2023), potentially indicating southern residents are spending more time elsewhere to locate Chinook salmon.

1.4 Shipping noise impacts to declining populations in the Northeast Pacific Ocean

Areas of high shipping traffic overlap with critical habitat for resident killer whales (Veirs et al., 2016; Williams and O'Hara, 2010) and migration corridors for Chinook salmon. Both resident killer whale (northern and southern) populations have been documented to alter their behaviour in response to shipping traffic (Foote et al., 2004; Holt and Johnston, 2011; Holt et al., 2009; Lusseau et al., 2009; Wieland et al., 2010; Williams et al., 2009, 2002). Resident killer whales display horizontal and vertical avoidance responses to increased boat traffic (Williams et al., 2009, 2002), as well as increased amplitude of vocalizations (Holt et al., 2009) and call duration with increased vessel activities (Foote et al., 2004). Resident killer whales have also been documented to switch away from feeding to traveling when the number of vessels in close proximity (<400m) increases, which could be displacing killer whales from their optimal foraging areas (Lusseau et al., 2009). Recently, dose response curves have been developed to set thresholds for sound pressure levels that will likely induce behavioural changes (Joy et al., 2019; Williams et al., 2014). Sound pressure levels of 129.5 dB re 1µPa have a 50% probability of causing resident killer whales to have a low behavioural response, while sound levels of 137.2 dB re 1µPa have a 50% probability of causing moderate behavioural responses (Joy et al., 2019).

Research into the impacts of shipping noise on fish and invertebrates is increasing, but no research has been conducted on Chinook salmon. One study has examined the impact of boat noise on juvenile pink (*Oncorhynchus gorbuscha*) and chum (*Oncorhynchus keta*) salmon, finding that at sound levels of 140 dB re 1µPa, 50% of salmon will behaviourally respond to the noise (van der Knaap et al., 2022). Juvenile pink and chum salmon have also been examined for behavioural changes from pile driving (Feist et al., 1992). Feist et al. (1992) found that pink and chum salmon demonstrated horizontal displacement due to the pile driving activities, but not vertical displacement. Changes in the size of the salmon school was also documented (Feist et al., 1992). These behavioural changes in response to underwater noise by other species of Pacific salmon indicate that Chinook salmon likely respond as well to increased underwater noise. Additionally, there has been little research on sound production or general behaviours/movements in wild adult Chinook salmon (Candy and Quinn, 1999; Courtney et al., 2021; Neproshin, 1972; Neproshin and Kulikova, 1975; Welch et al., 2013), representing key areas of research needed before understanding impacts from shipping noise.

1.5 Thesis Overview

My dissertation focuses on understanding additional noise sources to the marine soundscape that have not been previously explored, including noise from anchored commercial vessels and tugboats, as well as the potential acoustic (sound production) and behavioural impacts of these and other vessel noise sources on Chinook salmon and the consequences for apex predators, like killer whales. As Chinook salmon play a critical role in the Northeast Pacific Ocean ecosystem and shipping activities will only continue to increase, understanding how they are being impacted is crucial for conserving and protecting ecosystems in British Columbia. The research was all conducted on Vancouver Island, with a focus occurring in Cowichan Bay, British Columbia. Cowichan Bay Chinook salmon have been researched by Cowichan tribes, British Columbia Conservation Foundation and Fisheries and Oceans for many years, with escapement tracked since the 1980s and the establishment of a community hatchery in 1979

(Cross et. al., 1991; Tompkins et al., 2005), allowing for baseline understanding of migration timing, survival, and influence of anthropogenic factors.

The second chapter of my dissertation examines an unexplored anthropogenic noise source, the noise produced from commercial vessels while they are at anchor. Vessels in motion have been examined for many years, and mitigation measures are being considered to reduce the noise of vessel in motion (MacGillivray et al., 2019; Williams et al., 2019), but noise from anchored vessels has had a few studies examine their impact (González Correa et al., 2019; Ivanova et al., 2020; Rountree et al., 2020). SoundTrap underwater acoustic recorders (Ocean Instruments, New Zealand, system gain: -171.5 to -175.4 dB re FS/µPa, flat frequency response from 10 Hz to 72 kHz, self-noise < 34 dB re 1 µPa above 2 kHz) were deployed near anchorage locations in Cowichan Bay, British Columbia between August, and October 2019 and 2020. Sound pressure levels and frequency composition with and without an anchored vessel were compared for changes in noise levels. Significant changes in the underwater soundscape were observed when an anchored commercial vessel was present, elevating the underwater soundscape. These results highlight an underrepresented anthropogenic noise source that needs to be further explored to understand the impacts to marine species.

My third chapter explores another underrepresented anthropogenic noise source, tugboats, which have only been minimally explored (Amron et al., 2021; Houweling et al., 2022; Sprague et al., 2016). We deployed hydrophones at two locations on Vancouver Island, British Columbia (Cowichan Bay and Campbell River) where tugboats were consistently present. Similarly, to chapter 2, both sound pressure levels and frequency spectra were examined. At both locations the sound pressure levels were significantly elevated, but substantial changes to the frequency spectra was only observed in Cowichan Bay. We demonstrated that tugboats are significant contributors to the underwater soundscape but had a larger impact in more sheltered locations with less traffic from other types of vessels.

The fourth chapter of my dissertation examines sound production in three species of Pacific salmon (*Oncorhynchus* spp.), Chinook salmon, coho salmon (*Oncorhynchus kisutch*) and pink salmon. Research on sound production in Pacific salmon comes mainly from the 1970s (Neproshin, 1972; Neproshin and Kulikova, 1975; Kuznetsov, 2009), with little to no detailed descriptions. To be able to examine the impact of underwater noise on sounds produced by Pacific salmon, first detailed descriptions need to be outlined. Hydrophones were deployed at the Big Qualicum Hatchery on Vancouver Island, British Columbia in September, and October 2017. Hydrodynamic, air movement and unknown mechanism sounds were found for all three species, with differences in types of sounds, percent occurrence and sound frequencies observed. Results from this chapter demonstrate the range of sounds produced by some Pacific salmon species and is a starting place for examining the potential impacts underwater noise have on the sounds produced.

My final chapter (5) examines the movement and behaviour patterns of adult Chinook salmon prior to river entry, and the impact that underwater noise has on their behaviours. Minimal research has been conducted on adult Chinook salmon movements (Candy and Quinn, 1999; Welch et al., 2013), and no research to date has examined the impacts of underwater noise on Chinook salmon behaviour. To address these questions, adult Chinook salmon were acoustically tagged in Cowichan Bay, British Columbia in the fall of 2019, 2020, and 2022. Generalized additive models revealed consistent daily patterns in detections and behaviours (depth and activity) in Cowichan Bay. Depth values significantly increased with increased sound pressure levels and significant changes were observed with number of AIS vessels present for both depth and activity levels. This chapter is the first study to demonstrate the impacts of underwater noise on Chinook salmon behaviour.

All together my dissertation represents the first detailed examination of sound production as well as the general movements and behaviour of terminal Chinook salmon (prior to and during river migrations), and a first look at the impacts of underwater noise on some of these behaviours. Chinook salmon populations have seen declines in marine survival (Welch et al., 2021), size-at-age (Ohlberger et al., 2019), and fecundity (Malick et al., 2023) in the last few decades, leading to fewer individuals returning to spawn (Riddell et al., 2013), with many anthropogenic factors influencing their survival (Cao et al., 2022; Levin et al., 2001; Nehlsen et al., 1991). Previously, underwater noise had not been explored as a potential threat to Chinook salmon survival, but my dissertation demonstrates how increased underwater noise levels could be influencing Chinook salmon behaviours and potentially their survival.

Chapter 2 - Anchored bulk carriers have substantial impacts on the underwater soundscape in Cowichan Bay, British Columbia

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2.1 Abstract

In recent decades shipping traffic has increased, leading to elevated underwater ambient noise levels. Research has been conducted on the noise generated by ships underway, however little is known about potential noise from ships at anchor. In coastal regions, commercial vessels can seek anchorages prior to entering port, leading to concern regarding the impacts on the soundscape and marine ecosystems. Cowichan Bay, British Columbia, a coastal region (800 Ha) 70 km away from the Port of Vancouver, was examined as a case study to understand the possible soundscape contribution from anchored bulk carriers. When a carrier anchored, sound pressure levels (SPL: 20-24000 Hz) were elevated 2- 8 dB re 1 μPa throughout the bay. These results demonstrate the change anchored carriers can have on underwater soundscapes and is an important step in understanding the potential impact these vessels may have on marine organisms and important ecosystems.

2.2 Introduction

The number of global commercial shipping vessels has increased four-fold during the last 20 years (Tournadre, 2014), adding substantially to underwater ambient sound levels in low frequencies (< 1000 Hz; Ross, 1976; Veirs et al., 2016). Large bulk carriers and container ships produce sound at 184.2-188.1 dB re 1 μPa^2 (source level) at frequencies between 20-1000Hz with higher levels detected from the stern (McKenna et al., 2012). Individual commercial vessel passages can elevate sound levels (100 – 15,000 Hz) by as much as 20 dB above ambient levels (Veirs and Veirs, 2006). Additionally, the speed and size of commercial vessels influences the sound levels produced (Arveson and Vendittis, 2000; Ross, 1976). For container ships, vessel speed was the best predictor of source levels, but trends in the influence of total length and gross tonnage were also observed (McKenna et al., 2013). Research to date has focused on the impacts on the underwater soundscape from these vessels while moving, and noise additions from anchored commercial vessels awaiting access to port are often unaccounted for.

When carriers are in motion, most of the noise is from propeller cavitation, but with some noise generated by generators and other machinery onboard the vessel, especially at lower speeds (Arveson and Vendittis, 2000). When a carrier drops its anchor to the seafloor in order to remain stationary (anchoring), noise from propeller cavitation would be removed. Previous studies have documented elevated sound levels, and altered habitat use of marine species from recreational vessels (González Correa et al., 2019) and cruise ships (Ivanova et al., 2020) at anchor. Anchoring recreational vessels have also been documented to disrupt the physical benthic environment (Panigada et al., 2008). To our knowledge, no previous research has examined noise produced from bulk carriers at anchor, but likely noise produced from generators or other machinery would be produced while at anchor.

Passive acoustic monitoring is an important tool in evaluating the contribution of geophony, biophony and anthropophony to underwater soundscapes (Pijanowski et al., 2011), and has been previously used to quantify the influence of natural geophony (*e.g.*, wind and waves) and anthropogenic noise in the waters around Vancouver Island, British Columbia, also known as the Salish Sea (Burnham et al., 2021). A high presence of ships has been documented in the Salish Sea throughout the year (Erbe et al., 2012), and some of these commercial vessels are also anchoring in coastal regions around the Southern BC Coast with no time limit on how long they can anchor for (*Canada Shipping Act*, 2001). Here we consider the acoustic impact of anchored bulk carriers in Cowichan Bay, Vancouver Island, British Columbia as a case study of their potential influence on marine soundscapes. This is a highly industrialized site, frequently used by bulk carriers waiting for access to the Port of Vancouver, located approximately 70 km away.

2.3 Methods

2.3.1 Study location

Cowichan Bay, British Columbia, is located on the east coast of Vancouver Island at the mouth of the Cowichan River (48° 44.975' N, 123° 35.922' W). The bay is approximately 4 km long and 2 km wide (~ 800 Ha). Water depth increases gradually from 0 m at the head of the bay to approximately 70 m at the mouth (Figure 2.1). Within the bay there is one commercial anchorage site, with five more sites located outside extending southwards towards Saanich Inlet.

2.3.2 Passive acoustic recordings

Seven SoundTrap (300 STD/300 STDHF, Ocean Instruments, New Zealand) internally recording hydrophone systems were deployed throughout Cowichan Bay from 14 August, 2019 to 4 November, 2019, and then redeployed from 10 August, 2020 to 28 October, 2020 (Figure 2.1, Table 2.1). All SoundTraps were deployed in water approximately 15-45 m deep, with six deployed at the bottom. Five SoundTraps were deployed in shallow water (~20 m), and anchored to the shore using leaded line. Polysteel lines connected to 200 mm trawl floats were used to keep the hydrophones vertical in the water column at a distance of 1-2 m off the bottom. A sixth system was deployed in deeper (45 m; 1 m off bottom) water and was equipped with an acoustic release for retrieval. The final SoundTrap was the only one deployed from the surface and was secured to a leaded line that was connected to a 7 kg cannon ball and attached to a mooring buoy. This SoundTrap was deployed at a depth of 3 m from the surface, in 23 m of water. In 2020 an additional SoundTrap was deployed with an acoustic release outside Cowichan Bay from 12 August to 22 October in 25 m water depth (Figure 2.1).

All hydrophone systems sampled at 48 kHz at 16 bits. Two recorders, Front Mid and NS Open, were duty cycled, recording for 15 minutes every hour, while all others recorded continuously. The SoundTraps were calibrated by the manufacturer at 250 Hz using a B & K 2236 pistonphone at a source level of 120 dB re 1 µPa before being deployed. Data were stored as compressed sud files (.SUD) on internal memory of individual SoundTraps until retrieval at end of the study.

Data were decompressed and downloaded after SoundTrap retrieval, using SoundTrap Host software (Ocean Instruments, New Zealand), with subsequent analysis conducted using original code written in Python (version 3.7). Sound Pressure Level (SPL) time series were calculated for three frequency bands (100-1000 Hz, 7500-8500 Hz and 20-24000 Hz) to represent the soundscape at a low-frequency range (100-1000 Hz), a mid-frequency range previously shown to be a good indicator of wind generated noise (7500-8500 Hz: Burnham et al., 2021; Vagle et al., 1990) and broadband representing the full recording range of the hydrophones based on the selected sampling rate (20-24000 Hz). Additionally, the frequency band of 100-1000 Hz overlaps with the known hearing range of Chinook salmon, *Oncorhynchus tshawytscha* (Oxman et al., 2007), which were migrating back to the Cowichan River during our study period. The soundscape was described by comparing results from each of the recorder locations using 15-minute averaged data. Data were not filtered to remove any abiotic, biotic, or anthropogenic (recreational boats, shore noise) influences, these were occurring everyday throughout our study so were assumed to be a constant. Biotic noise sources could originate from resident species of invertebrates, fish or marine mammals, but no vocalizing species of marine mammals (*e.g.*, humpback whales (*Megaptera novaeangliae*), southern resident killer whales (*Orcinus orca*)) were observed during the sampling period.

2.3.3 Anchored vessel analysis

All internationally travelling vessels over 300 gross tonnage and domestic travelling vessels over 500 gross tonnage are required to report position and identifying information using Automatic Identification System (AIS) during their passage and while at anchor (IMO, 2015). AIS position data (± 1 m) were obtained for every vessel equipped with AIS in 5-minute bins for an approximate 5 km radius around Cowichan Bay during August through October of 2019 and 2020. These data for each bulk carrier to enter Cowichan Bay were used to track each vessel during their arrival, at-anchorage and departure. Sound pressure level data from each station were then combined into two time periods: (1) no vessel and (2) vessel anchored. The 'no vessel' time period was defined as when no vessel was within 500 m of the anchorage location (yellow star in Figure 2.1) inside Cowichan Bay, and once a vessel was within 500 m of its anchorage (area within dashed circle in Figure 2.1) the period was labelled as 'vessel anchored'. The 'vessel anchored' time period was further broken down into vessel arriving, vessel at anchor (stationary, with anchor on bottom) and vessel departing at only the closest hydrophone mooring location, Genoa Bay. The arrival period was defined by a vessel being within 500 m of the anchorage location to the end of the first day. The beginning of the first full day at anchor to the end of last full day at anchor was defined as at-anchor, and the start of the final day to the time when the vessel reached 500 m from the anchorage was defined as departing.

Power spectral density (PSD) shows the power (dB) at each frequency of a given sound source and is a conventional way to present temporal and frequency dependent variation in a given soundscape (*e.g.*, Merchant et al., 2015). PSDs were calculated for each bulk carrier over a consecutive three-day period (24-hours prior to vessel arriving (no vessel); 24-hours with vessel arriving at hour 12 (arriving); and first 24-hours of the vessel at anchor (anchor)) using 1/3 octave bands at the 5th, 50th, and 95th percentiles. However, the first vessel of 2020 (Vessel 2, Table 2.2) arrived prior to the SoundTraps being deployed, so it was not included in this analysis.

The PSDs from each arriving and at-anchor vessel were subtracted from their respective no-vessel PSDs sound level changes for the entire frequency range collected (20- 24,000 Hz). We also calculated the empirical probability densities, which presents the full range of observations in the form of normalized histograms (Merchant et al., 2015), computed from minute-by-minute averages of the PSD. Here we calculated over two one-week periods in September 2019, representing periods without and with an anchored bulk carrier present. Diel comparisons were made, whereby the period between midnight and 05:00 (PDT), represented night-time conditions and the period between 11:00 and 16:00 (PDT) to represented daytime conditions.

Positional information was used to define the circle the vessel made around the anchor spot as a result of the changing tide in the bay. The AIS information established the exact anchor location, which was used with the vessel motion to investigate the directional variability of the received noise field. Vessel 1, anchored in the bay in 2019, was used as an example vessel and was the only vessel analyzed for positional information.

Comparisons of median sound pressure levels between times when a bulk carrier was present versus times when no carrier was at anchor, and between the different time periods for individual carriers (Arrive, Anchor, Depart) were not normally distributed, and therefore non-parametric Mann-Whitney-Wilcoxon t Tests and Kruskal–Wallis ANOVAs with Dunn's post hoc tests were used for analysis. All statistical tests were performed in RStudio version 4.1.2.

2.4 Results

2.4.1 Soundscape description

Acoustic recordings were collected for between eight to ten weeks in 2019 and 2020 (depending on hydrophone), except for one location (marina) in 2019, which was missing a few days in the middle of the deployment, resulting in approximately 7 weeks of recordings. Cowichan Bay has a high level of anthropogenic noise that originates from recreational boats entering and leaving two marinas, and from commercial activity on shore. The soundscape of the bay exhibited a strong diel pattern both in 2019 and 2020 that was broadband in nature but showed the strongest pattern at frequencies between 7500 and 8500 Hz (Figure 2.2). Median (5%, 95% CI) sound levels increased to 76.5 dB re 1 µPa (71.0, 93.0 dB) starting at 06:00 (PDT) each day and peaked at a level of 87.2 dB re 1 µPa (76.5, 101.5 dB) at 13:00 (PDT) before dropping to 76.3 dB re 1 µPa (71.3; 93.3 dB) at 20:00 (PDT). Overnight (21:00-05:00) levels ranged between 74.7 dB re 1 µPa (70.2, 84.5 dB; 0200) and 75.2 dB re 1 µPa (70.7,87.0 dB;

2100) depending on the hour. This pattern was detected throughout the study area in both years without any observable shifts in the onset time and duration. The source of this sound increase is yet to be fully determined.

2.4.2 Anchored vessel analysis

The anchorage location inside Cowichan Bay (Figure 2.1) was used by bulk carriers in both years. In 2019, during the first 25 days of the study no vessels anchored until 8 September when one arrived and stayed for 18.5 days (Table 2.2). During the remaining 27 days of the study no bulk carrier was present. In contrast, in 2020 there were only 25 days when a bulk carrier was not anchored, with four different bulk carriers anchoring between 12 August and 30 October. These vessels stayed at anchor between 2 and 22 days. All bulk carriers for both years were of similar size (200-329 m total length), but with slight differences in reported tonnage (35800 - 43300 gross tonnage; Table 2.2).

Longer-term broadband timeseries showed the bulk carriers' acoustic impact on the bay. Broadband sound pressure levels measured at the closest SoundTrap (Genoa Bay: 700 m) from the anchorage location increased significantly from a median value of 101 dB re 1 µPa (93, 115 dB; 5%, 95% CI) when no carriers were anchored, to 109 dB re 1 µPa (103; 117 dB) when a single carrier was anchored (Mann-Whitney-Wilcoxon t Test, $p < 0.0001$, $W = 7261392$). The increase in sound pressure levels were observed for the entire time of a carrier being at anchor (Figure A2.1), which varied from 2- 22 days (Table 2.2). Additionally, the increase in sound pressure levels with an anchored vessel was significantly elevated at all stations (2-8 dB increase: Mann-Whitney-Wilcoxon t Test, $p < 0.0001$) located inside Cowichan Bay but was not observed at the Sep Point station (~1 dB increase: Mann-Whitney-Wilcoxon t Test, $p = 0.244$, $W = 3820744$) which was the only station located outside the bay (Figures 2.1, 2.3).

Median power spectral density plots showed bimodal changes in the frequency composition when a bulk carrier was arriving in the bay and while it was at anchor (Figure 2.4). Frequencies most impacted (> 5 dB re 1 µPa median difference) were observed at < 100 Hz and between 1000-5000 Hz, specifically in the 5th and 50th percentiles. The 95th percentiles were the least impacted by an anchored bulk carrier for the full frequency range but were still impacted at frequencies below 100 Hz.

The Genoa Bay SoundTrap data were used for empirical probability density analysis (Figure 2.5). The nighttime periods between midnight and 05:00 (PDT) with no bulk carrier anchored in the bay were relatively quiet at all frequencies (Figures 2.5(a) and 2.6) with the spread of PSD values less than 30 dB re 1 µPa. At frequencies above 3 kHz the data were constrained by the noise floor of the instrument. During daytime hours in the absence of any anchored vessel the variability increased significantly, and the maximum to minimum difference in observed PSDs increased to 60 dB (Figure 2.5c), presumably due to increased boating activity in the bay combined with the unexplained daytime increase in noise levels shown in Figure 2.2 (Figure 2.5c). At frequencies above 3 kHz there is a secondary modal ridge approximately 5 dB above the sensitivity level of the instrument, presumably as a result of this unexplained noise source. Also, at frequencies below 60 Hz, during periods with no anchored vessel present, the PSDs are lower than the sensitivity of the instrument and therefore not detectable. When a bulk carrier was anchored in the bay the PSDs and empirical probability densities changed significantly (Figure 2.5b, d). Except for at frequencies below 20 Hz, the observed noise levels did not go below the sensitivity of the instrument and there were minimal differences between daytime and nighttime. The vessel noise also masked the unexplained daytime noise source. The PSDs when a carrier was anchored showed a change in the frequency composition. A broad peak centered around 1 kHz, but additionally a number of harmonics and bimodal structure between about 30 Hz and 300 Hz were observed (Figures 2.5-2.6).

Individual bulk carriers also had different effects on the soundscape while arriving, at anchor, and when departing Cowichan Bay (Figure 2.7). The largest bulk carrier (vessel 1) by gross tonnage was anchored in 2019 and vessel 4 was the smallest (anchored in 2020), both anchored the longest over the 2 years (Table 2.2). When vessel 1 was arriving median (5%, 95% CI) sound pressure levels were 112 dB re 1 µPa (106; 121 dB; 95 CI), which was significantly elevated compared to the levels observed when the smaller vessel 4 arrived (median sound pressure levels of 108 dB re 1 µPa (104; 115 dB)) (Kruskal-Wallis, $p < 0.0001$, df = 3). A similar trend was observed while vessel 1 was at anchor, where median sound pressure levels were 112 dB re 1 µPa (104; 118 dB), compared to when vessel 4 was at anchor and the median sound pressure levels were 107 dB re 1 µPa (103; 113 dB) (Kruskal-Wallis, $p < 0.0001$, df = 3). Lastly, vessel 1 also had higher median sound pressure levels when departing (112 dB re 1 µPa (103; 120 dB), as compared to when vessel 4 departed (105 dB re 1 µPa (103; 116 dB) (Kruskal-Wallis, $p < 0.0001$, df = 3).

2.4.3 Directionality of noise from an anchored bulk carrier

The AIS data from vessel 1 (Table 2.2) at anchor in Cowichan Bay between 8 and 26 September 2019 were used to determine the actual anchor location from positional variations as the vessel rotated due to tidal forcing (Figure 2.8a). This location was 632 m away from the Genoa Bay SoundTrap mooring. As the vessel spun around the anchor the observed SPL in a frequency range of 0.01-20 kHz varied by as much as 7.5 dB re 1 µPa as the vessel turned, with the highest noise levels when the bow was towards the hydrophone, and lowest levels when the stern was pointed towards the same hydrophone (Figures 2.8b, c). A port-starboard asymmetry was also seen for this vessel, with no significant differences between the bow towards the hydrophone and starboard side of the vessel towards the same hydrophone, while a reduction of several dBs was noted when the port side was toward the mooring. Frequency dependent changes were also observed between different headings, with the largest difference detected at frequencies below 60 Hz (Figure 2.8d-f). Frequencies below 60 Hz were lower when the bow or stern of the vessel was pointed towards the hydrophone compared to non-head-on headings (90° or 270°).

2.5 Discussion

Our soundscape analysis of Cowichan Bay showed a strong diel pattern in sound levels. Sound levels between 7500 and 8500 Hz were elevated during daytime hours (06:00-20:00 PST), with this unaltered with changing daylength during our study period. This pattern was also not connected to tidal or wind patterns for the area during this period (Figure A2.2 and A2.3). Additionally, since the consistent pattern was observed throughout the bay and not just by marinas or high boat traffic, we assume it to be from a biological source. One potential option is snapping shrimp (*Betaeus* spp.). Other species (*Alpheus* spp. and *Synalpheus* spp.) have been observed to increase their snap rate during the day compared to overnight (Lillis and Mooney, 2018) and individual *Synalpheus paraneomeris* can produce sounds as loud as 190 dB re 1 µPa (Au and Banks, 1998). Snapping shrimp residing in Cowichan Bay could be the cause of the strong diel pattern observed in our study, but further investigation would be required to confirm.

Our data from Cowichan Bay showed anchored bulk carriers can have substantial impacts to the underwater soundscape in coastal marine systems. Large ships like bulk carriers anchoring in coastal habitats can generate extensive underwater noise that is detectable at least 2 km away at the furthest station from the anchorage location (SS Open: Figure 2.1), but could potentially be detected further. Change was most detectable using the 5th percentiles, while the 95th percentiles were less impacted, indicating that anchoring bulk carriers are reducing the amount of time at lower sound pressure levels (< 95 dB) in the bay, but not elevating maximum sound pressure levels observed.

Diesel generators aboard commercial vessels produce tonal harmonics, with harmonics at 24 and 30 Hz detected even when the vessel is in motion and are independent of vessel speed (Arveson and Vendittis, 2000). Harmonics at 24 and 30 Hz were visible in the spectral probability density (Figure 2.6) while a bulk carrier was anchored in Cowichan Bay and could be generated from the generators aboard these carriers. Additionally, AC power lines to other machinery on these vessels can produced harmonics of 60 Hz (Arveson and Vendittis, 2000), and could be contributing to the noise produced. Finally, the anchor and attaching chain could be producing sounds, but more research would be needed to confirm these noise sources.

The analysis clearly showed that the noise field emanating from an anchored bulk carrier is highly directional. Previous research demonstrates that when vessels are traveling, higher sound pressure levels are detected originating from the stern (McKenna et al., 2012). However, in our study we found the opposite. While at anchor the propulsion machinery and propeller(s) are stopped, so other noise sources clearly dominate the noise generated by the ship. It is therefore expected that the directional characteristics of this noise will be different than when the vessel is underway. Future research could aid in examining the specific origin of these directional changes in sound levels and frequency composition.

In the waters around Vancouver Island, anchored bulk carriers are common in areas like Cowichan Bay, for example, 350 carriers were recorded to be anchored in the southern portion of Vancouver Island (Victoria to Nanaimo BC; 45 total locations) in 2019, and 606 anchored in

2020. Some are anchored for more than two weeks as was observed in this study, adding to the soundscape for extended periods (Figure A2.1). Cowichan Bay anchorage locations represent ~13% (6/45) of the anchorage locations in Southern Vancouver Island but can also accommodate larger vessels (>220 m length) potentially leading to more demand for anchoring in Cowichan Bay (Transport Canada, unpublished data). Between 2019 and 2020 the six anchorages in Cowichan Bay accommodated 15%-20% of the yearly anchorages of bulk carriers.

Additionally, the use of anchorage locations varies year to year but appears to have been impacted by the COVID-19 pandemic. For example, the six anchorage locations in and around Cowichan Bay had between 5 and 9 carriers anchored during August-October of 2016/2018/2019, while in 2020 there were 16 carriers that were anchored during the same period. The use of the anchorages around Cowichan Bay appears to have resumed to normal levels in 2021 when 10 carriers were anchored during August and October. Marine systems have been documented to have been quieter during the global shutdown of shipping activities in 2020 (Bates et al., 2021; Thomson and Barclay, 2020), however, bulk carriers could not come into ports and were restricted to anchoring in coastal waters for extended periods of time.

Cowichan Bay hosts many fish and invertebrate species, and forms part of the key migration corridor for Pacific salmon (*Oncorhynchus* spp.). Fish exhibit changes in behaviour in the presence of elevated noise levels, with reduced foraging effort and success observed in three-spined stickleback (*Gasterosteus aculeatus*) and European minnow (*Phoxinus phoxinus*) while exposed to noise from a single boat passage (Voellmy et al., 2014a). Ivanova et al. (2020) noted horizontal displacement and changes in behaviour patterns of tagged Arctic cod (*Boreogadus saida*) while cruise ships were anchored and moving into a bay in the Arctic. In addition, a stress response has been documented for fish and invertebrate species subject to vessel noise (Celi et al., 2015; Wysocki et al., 2006). Results from this study demonstrate a further source of human activity that could alter their habitat quality.

Anchored bulk carriers in Cowichan Bay demonstrated a significant alteration to the underwater soundscape, however, our study is not without some limitations. Our study only examined one anchoring location in Cowichan Bay that is the furthest away from the next closest anchorage (2 km), which is not seen in other locations around British Columbia or the globe where multiple vessels are anchored at the same time in close proximity (~1 km). Future research should examine the influence of multiple anchored carriers to examine how the underwater soundscape would be altered with more anchored vessels. Additionally, only one type of commercial vessel was examined, it is unknown how different vessel types (*e.g.*, container ships) might influence the soundscape. Differences in spectral characteristics have been observed while different types of commercial vessels are moving (McKenna et al., 2012), so it is likely differences could be observed while at anchor. Lastly, our study was conducted in a shallow (<60 m water depth), sheltered bay over a two-month window. Seasonal oceanic conditions have been documented to influence noise produced by container ships, with higher source levels observed during the spring (April/May) compared to summer or fall (McKenna et al., 2013). This difference was potentially linked to warmer water at the surface in summer and fall that can trap sound waves (Jensen et al., 2011). Our study was conducted between August and October with warmer surface temperatures, which could reduce the noise received at our hydrophones creating an underestimation of the noise produced by the anchored carriers. Future research should examine the anchorage of different types of vessels, at different maximum depths and bathymetries over extended periods of time (year) to aid in understanding the impact that these carriers might have on a larger scale.

The global shipping industry has been changing the ambient underwater soundscape for the past few decades (reviewed in Hildebrand, 2009), with noise levels in the Northeastern Pacific Ocean increasing by ~3 dB per decade (McDonald et al., 2006). However, anchoring of these vessels prior to going into ports is minimally accounted for in the literature. Our study demonstrates the substantial impact anchored bulk carriers have on the underwater soundscape and represents a starting point to explore their impact further.

Table 2. 1. SoundTrap moorings and Cowichan Bay vessel anchorage information.

Location	Latitude (N)	Longitude (W)	Water depth (m)	Hydrophone depth (m)
2A	48.7589	123.6124	19	18
Front Mid	48.7523	123.6194	45	44
Marina	48.7455	123.6221	23	3
Genoa Bay	48.7556	123.5987	19	18
SS Mid	48.7394	123.6054	22	20
NS Open	48.7450	123.5755	18	17
Sep Point	48.7456	123.5689	25	24
SS Open	48.7334	123.5832	15	13
Anchorage location	48.7495	123.5987	58	NA

Table 2. 2. Specifications for bulk carriers anchored inside Cowichan Bay in 2019 and 2020.

Year	Name	Length (m)	Breadth (m)	Gross Tonnage	Length of stay (days)
2019	Vessel 1	329	32	43300	18.5
2020	Vessel 2	225	32	40000	10.9
2020	Vessel 3	200	36	38200	15.4
2020	Vessel 4	200	32	35800	22.2
2020	Vessel 5	225	32	40100	2.7

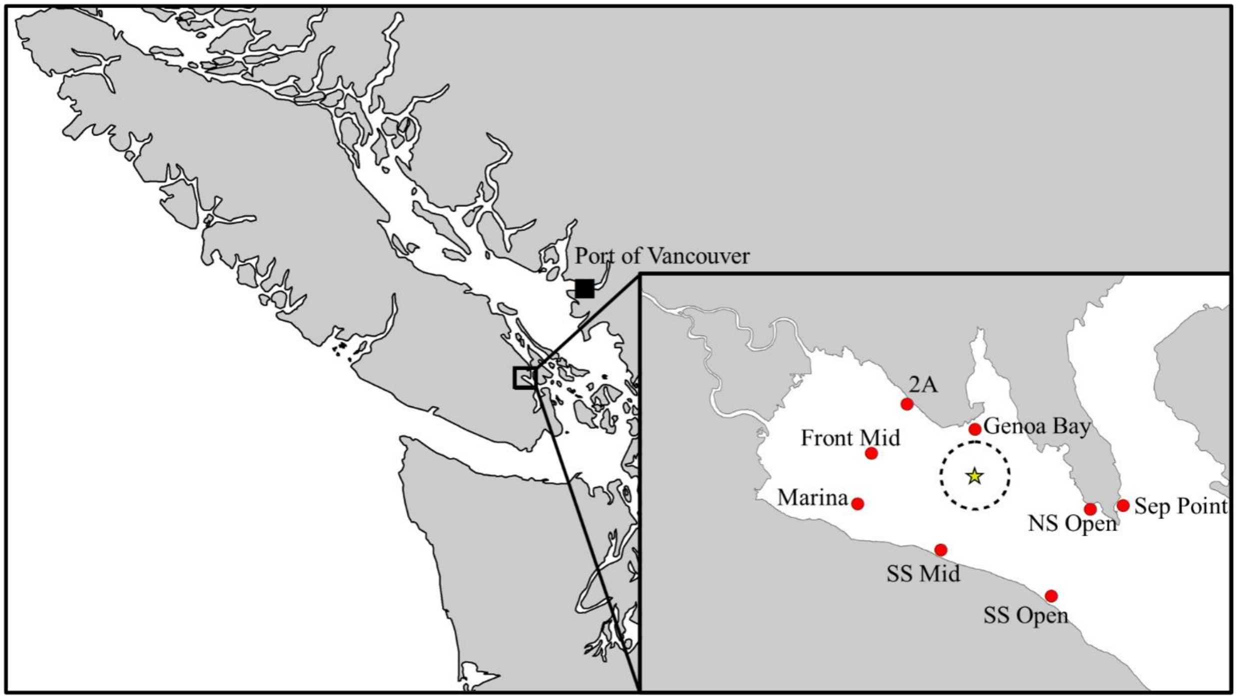

Figure 2. 1. Map of Vancouver Island with portions of lower British Columbia and Western Washington State. Black box denotes location of the Port of Vancouver. Inlet: Map of Cowichan Bay, BC. Red circles show SoundTrap hydrophone mooring locations around the bay and the yellow star denotes a bulk carrier anchorage location. Black dashed circle denotes 500 m radius around anchorage location. Map provided from Natural Earth and BC Provincial data catalog.

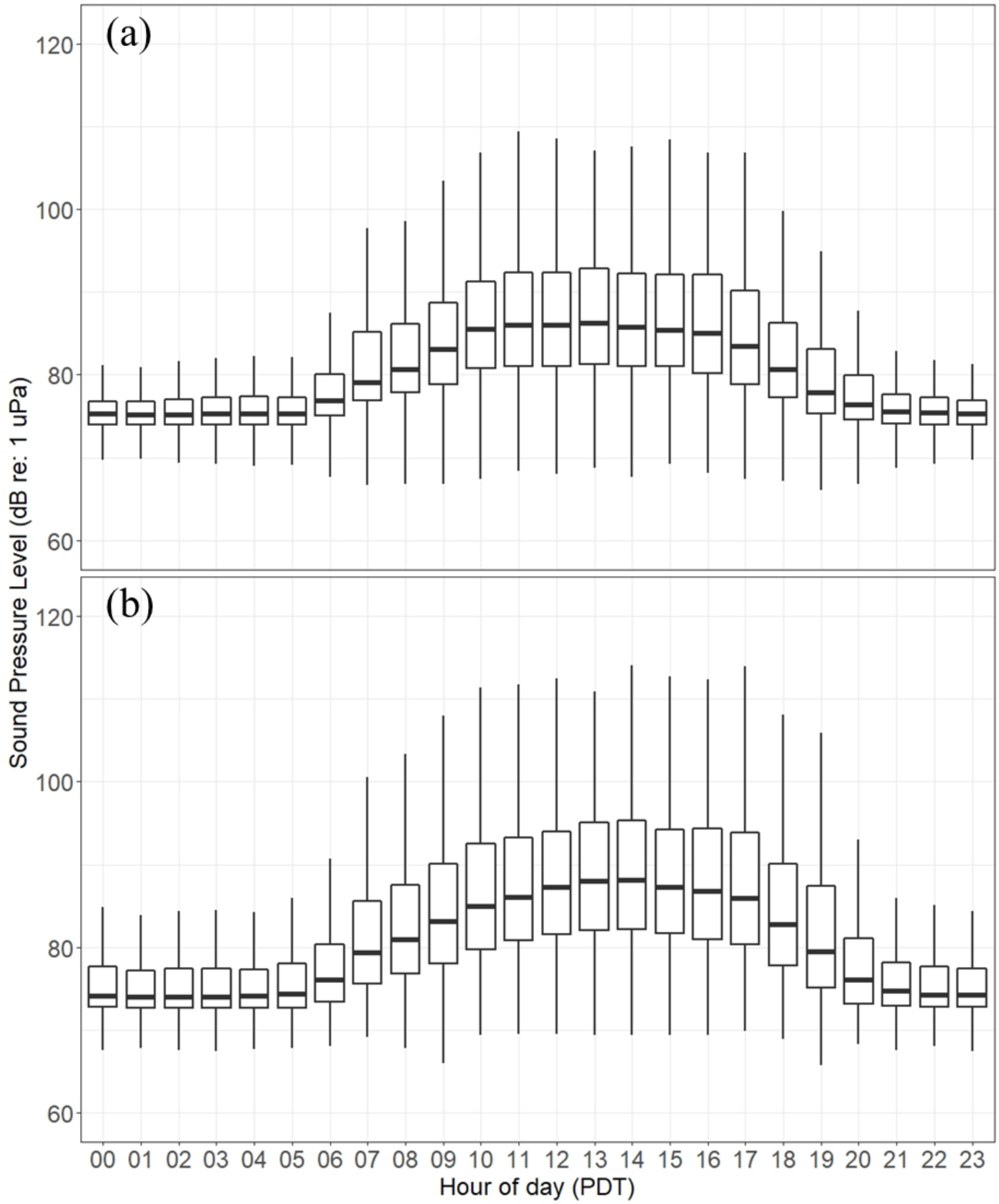

Figure 2. 2. Boxplots representing the diel pattern of sound pressure levels in the 7500-8500 Hz range using data from 2019 (a) and 2020 (b) from all hydrophones. Hour of the day shown in local time (PDT).

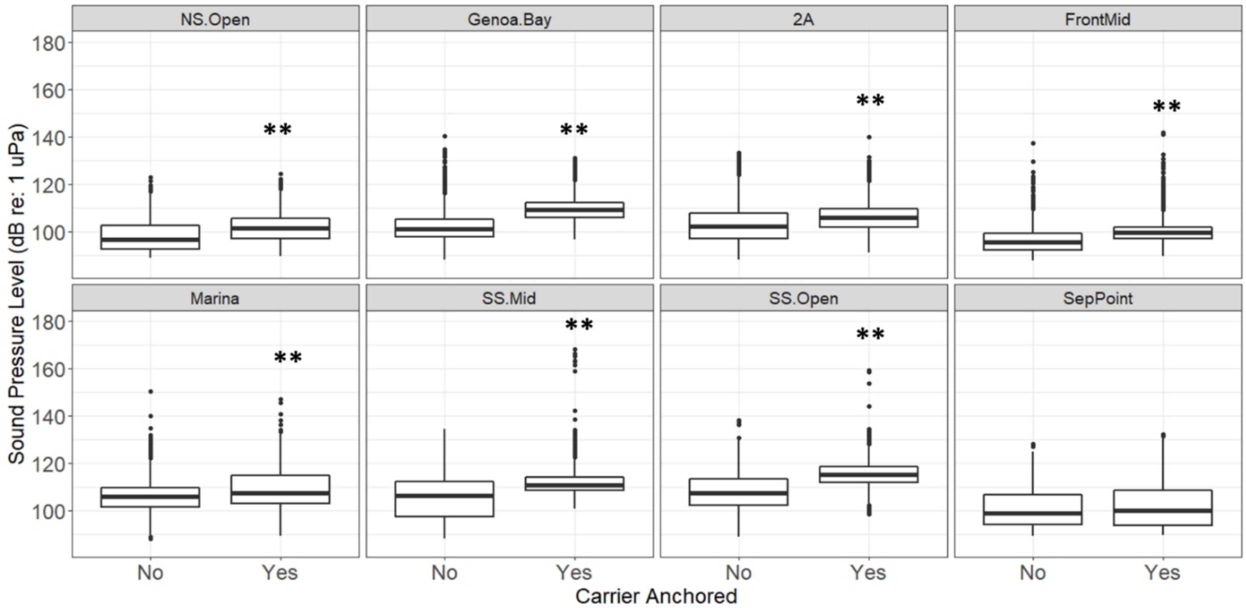

Figure 2. 3. Boxplots of sound pressure levels (broadband: 20-24000 Hz) at each station in Cowichan Bay, with data from both years combined. Median sound pressure levels increased when a bulk carrier was anchored at all stations except SepPoint. Asterisks indicate significantly different ($p < 0.0001$) sound pressure levels using Mann-Whitney-Wilcoxon t Tests.

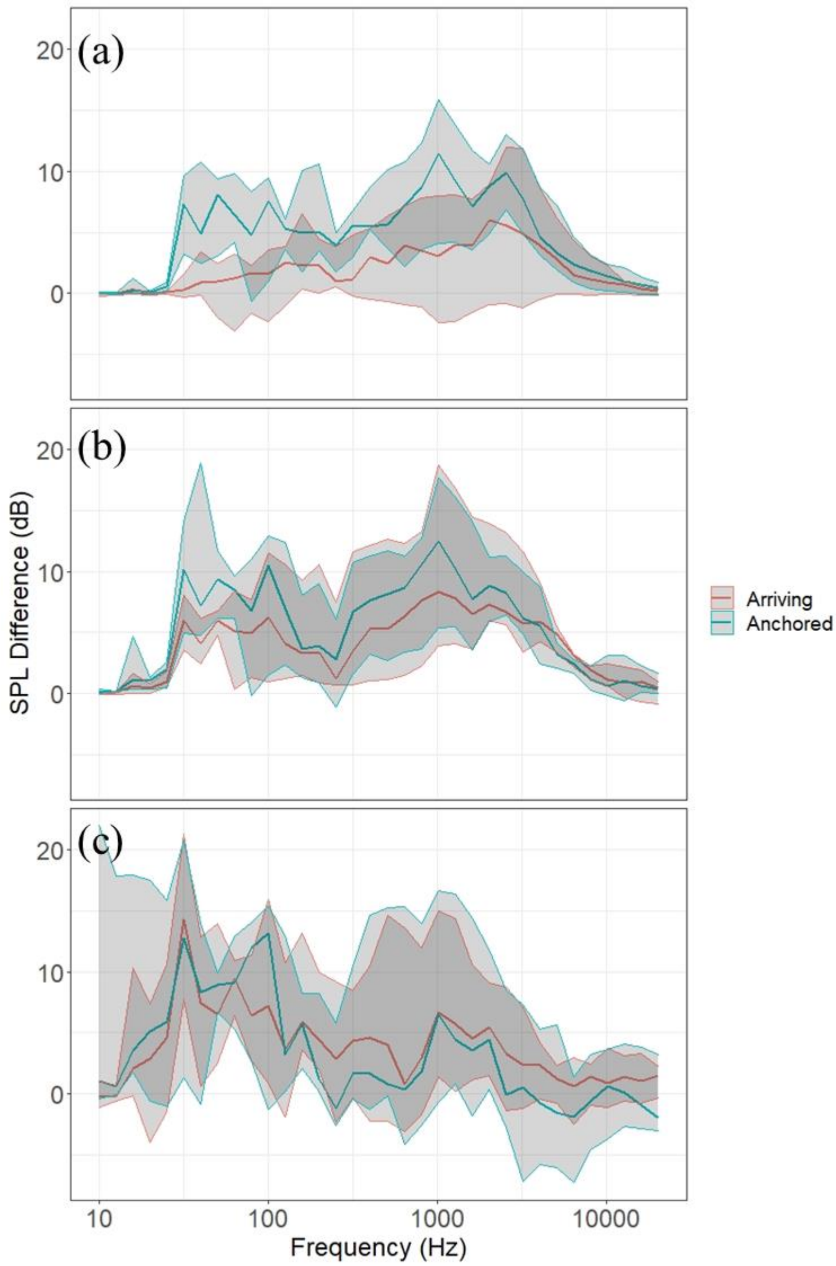

Figure 2. 4. SPL differences between when no bulk carriers were in Cowichan Bay and when bulk carriers were arriving (red lines) and anchored (blue lines) for 5th(a), 50th (b), and 95th (c) percentiles as functions of frequency. Shaded areas represent 5th and 95th confidence intervals around the median for each 1/3 octave band for the full frequency range of the recorder (20-24000 Hz).

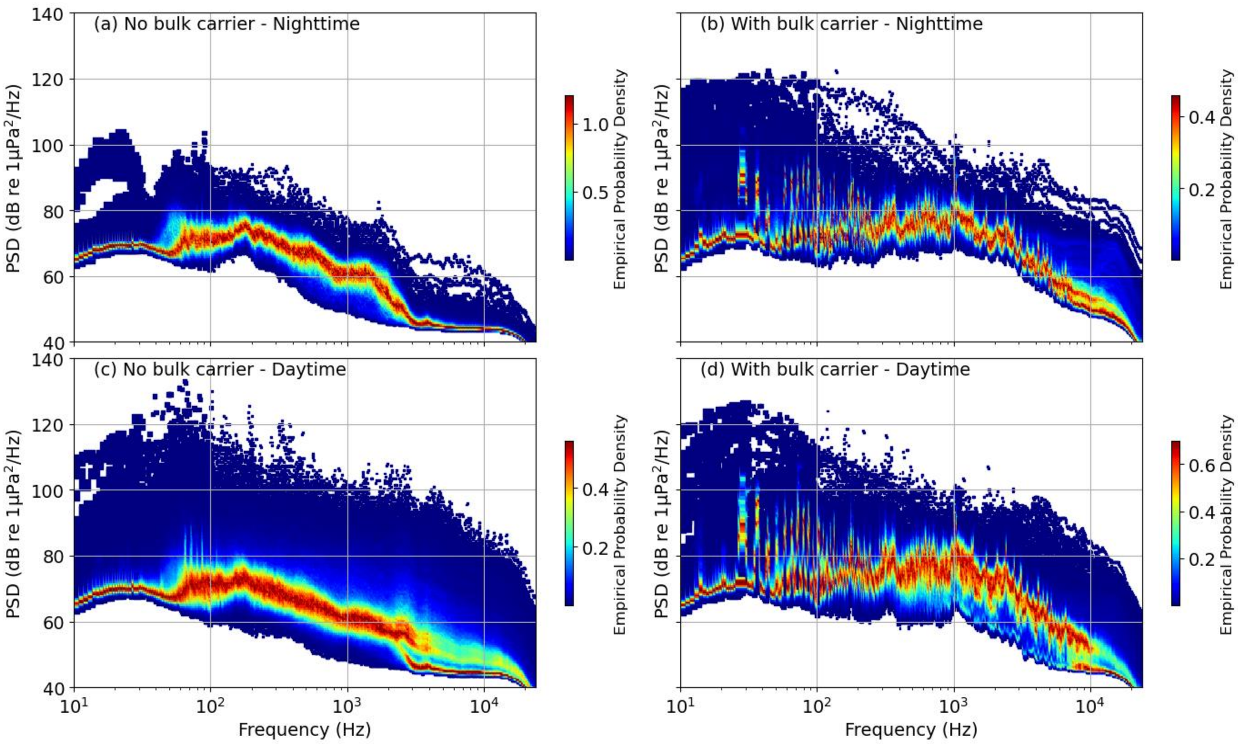

Figure 2. 5. Spectral probability densities of the Genoa Bay deployment for two 7-day periods without an anchored bulk carrier (1 September – 8 September, 2019) ((a) and (c)) and with a vessel present (10 September – 18 September, 2019) ((b) and (d)). Due to the significant day to night difference observed in the bay, the data have been further separated into midnight to 5 ((a) and (b)) and into 11 to 16 in the afternoon ((c) and (d)).

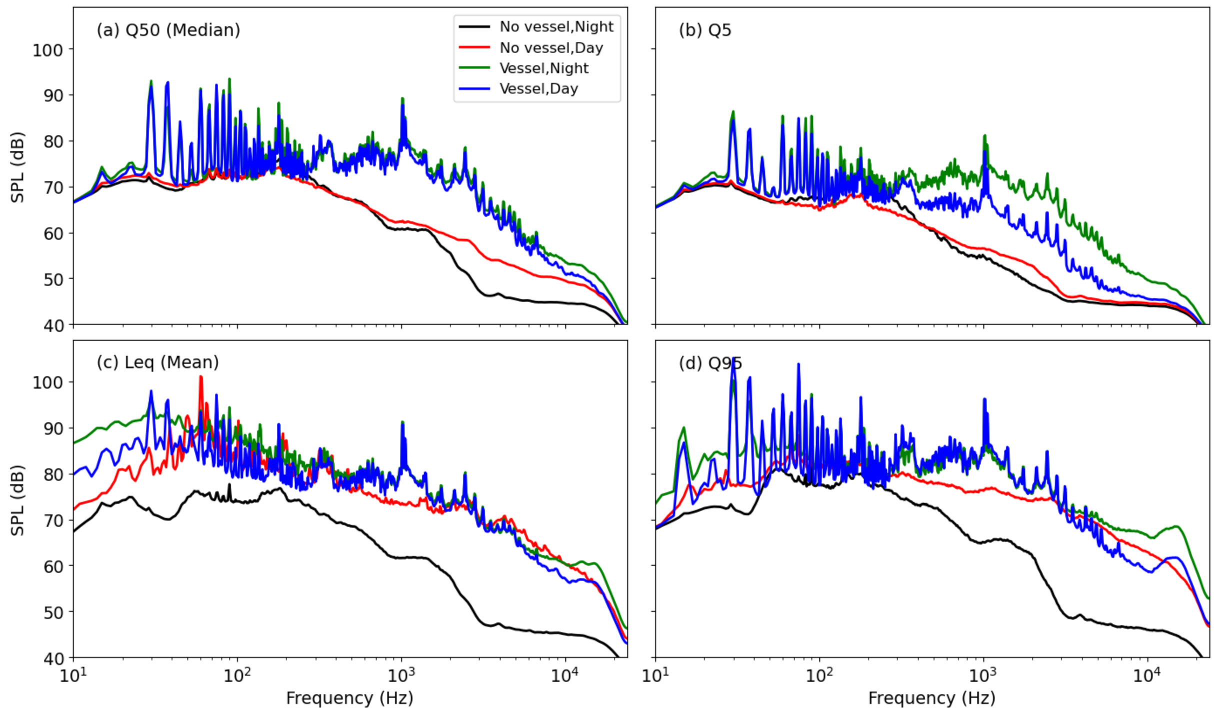

Figure 2. 6. Metrics extracted from the spectral probability density data shown in Figure 2.5 for the two one-week periods without and with a bulk carrier present and calculated for the daytime and nighttime periods. (a) observed median values (50th percentile), (b) 5th percentile, (c) mean values (Leq) and (d) 95th percentile.

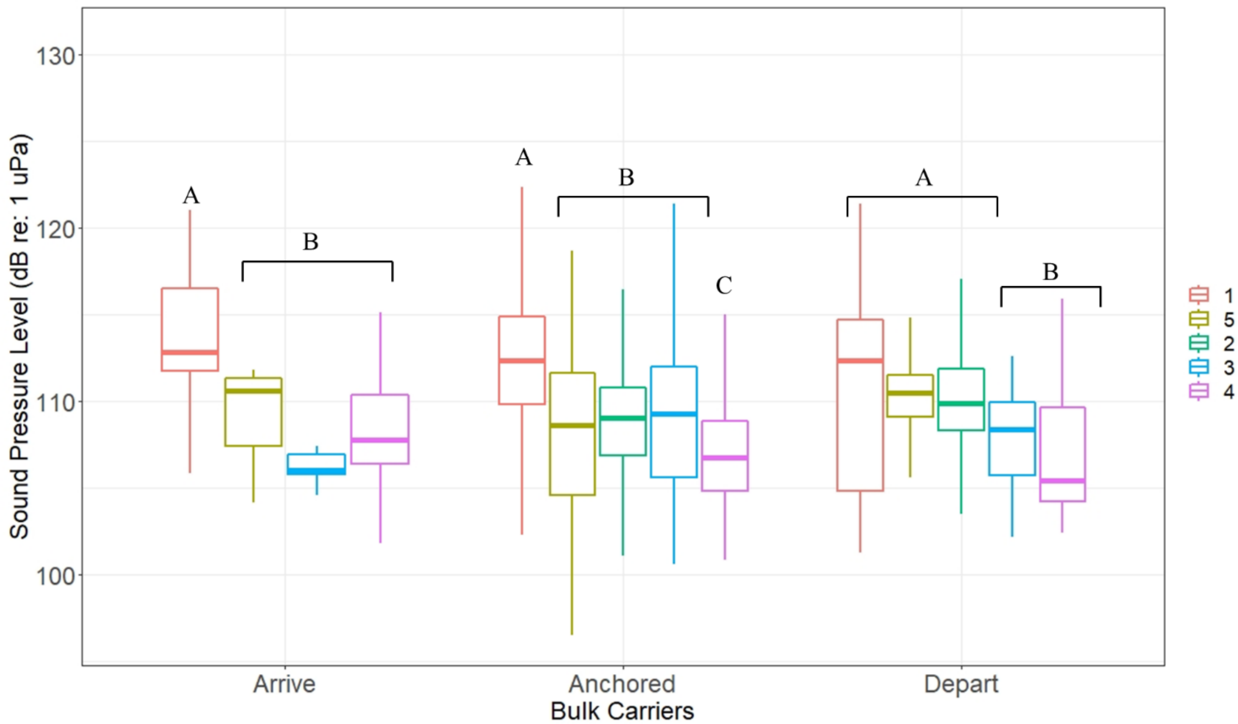

Figure 2. 7. Box plots of Sound Pressure Levels (broadband: 20-24000 Hz) for each of five bulk carriers arriving, at anchor, and departing Cowichan Bay. Letters indicate significantly different sound pressure levels within a time point using Kruskal-Wallis One-way ANOVA. Bulk carrier IDs are defined in Table 2.2 and ranked from largest to smallest by gross tonnage. No arrival data for vessel 2 as that carrier arrived before our recorders were deployed in 2020.

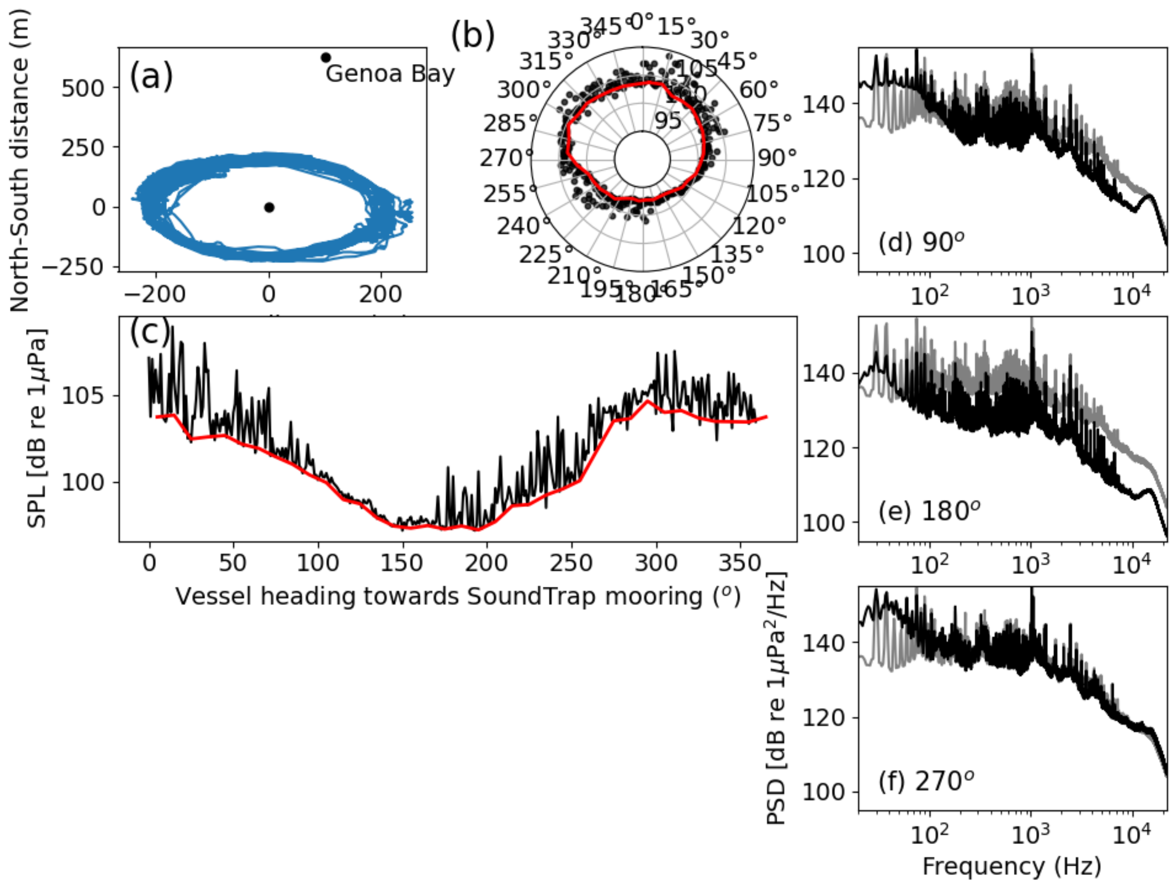

Figure 2. 8. Vessel noise directionality from vessel 1 (Table 2.2) anchored in Cowichan Bay between 8 and 26 September 2019. (a) AIS track and locations of anchor and Genoa Bay hydrophone mooring. (b and c) Polar and x-y plots of SPL (0.01-20 kHz) (black) as received by the Genoa Bay SoundTrap 632 m north. Red lines show the 10th percentile every 10 degrees in compass direction. (d)(e)(f) PSD at three different angles relative to the vessel anchor-SoundTrap direction. The PSD at 0° has been included in each figure as a reference (grey lines).

Chapter 3- Influence of tugboats on the underwater soundscape along the West Coast of Canada

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3.1 Abstract

Ambient underwater noise levels have been increasing, leading to concern about the impacts to marine species. Extensive research has been conducted on the changes to ambient noise levels of large commercial vessels, and potential mitigation measures. However, little is known about smaller commercial vessels (*e.g.*, tugboats) moving throughout coastal waters and what can be done to decrease their influence on marine species. To understand changes to the ambient noise levels (20-24,000 Hz) when tugboats are present, stationary hydrophones were deployed between August and October at two locations: Cowichan Bay, British Columbia (BC) (2019/2020) and Argonaut Wharf, Campbell River, BC (2021). Log transportation tugboats were assessed in Cowichan Bay and the results show significantly elevated median sound pressure levels (10 dB) when log transport tugs were detected near the estuary where log storage occurs. Even though Argonaut Wharf was used as a site without log transportation tugboats, tugboats were present throughout the study period. Median sound pressure levels were also significantly elevated (6 dB) when tugboats were present in Argonaut Wharf. The results demonstrate the impact that tugboat activities have on underwater soundscapes and are the first step in understanding the impact these activities have on marine organisms and important ecosystems.

3.2 Introduction

Anthropogenic noise has been modifying the global soundscape causing ambient levels to increase (reviewed in Hildebrand, 2009). One example is seen in the Northeast Pacific Ocean, where noise levels have increased by around 3 dB/decade for decades leading up to 2000 (McDonald et al., 2006). One main contributor to this increase is the shipping industry, which has been growing for many decades (Tournadre, 2014). Shipping noise dominates low frequency noise (40- 1000 Hz) which has increased by at least 10-12 dB since the 1950s in some areas (Ainslie et al., 2021; Ross, 1976). Noise produced by large commercial vessels has been extensively documented in the literature (*e.g.*, Gassmann et al., 2017; McKenna et al., 2013, 2012) and mitigation methods are being developed to reduce their impact on the underwater soundscape (Burnham et al., 2021; MacGillivray et al., 2019; Williams et al., 2019).

One shipping noise source that has only been minimally accounted for in the literature is noise produced by tugboats. Tugboats are used in coastal regions to push or tow equipment and to guide larger ships. Previous research demonstrated tugboats produce noise below 3 kHz (frequency range: 170-2,629 Hz) with a source level of 119 dB re 1 µPa (predicted) when transiting (not pulling or pushing) using a stationary hydrophone (Amron et al., 2021). When pushing a barge, Sprague et al. (2016) found that a tugboat produced noise at sound pressure level (peak) of 124 dB re 1 µPa (frequency range: 10-22,050 Hz) and a sound exposure level (received sound level over specified time) of 143 dB re 1 µPa (time constant = 0.125 s) at roughly 600 m away from a stationary hydrophone. They also documented that the frequencies most impacted by tugboat sound are frequencies below 1000 Hz. Vessel size has also been shown to have an impact on the noise produced, with tugboats over 50 m having a mean broadband radiated noise level (RNL; 10-64,000 Hz) of 186.2 dB re 1 μPa @ 1m compared to tugs less than 50 m that have a mean RNL of 180.5 dB re 1 μPa @ 1m (Houweling et al., 2022).

Around Vancouver Island, British Columbia, tugboats are operating in every month of the year (Houweling et al., 2022) and have the highest vessel hours detected compared to all other AIS equipped vessels in the area due to high volume of activity (MacGillivray et al., 2016). In addition to traditional roles, tugboats around Vancouver Island are also used to move logs into bays and estuaries where logs are stored prior to processing at pulp and sawmills. Logging booms have been documented to negatively impact estuarine habitats (Levy et al., 1990; Nahirnick et al., 2020; Power, 1987) but the additional noise generated by these activities has not been quantified. The goal of this chapter was to examine the underwater noise produced by tugboat activities in two different locations on Vancouver Island, British Columbia, to document differences in sound pressure levels and frequency composition under the different activities that tugboats are used for.

3.3 Methods

3.3.1 Study locations

Cowichan Bay, British Columbia, is an approximately 800 ha industrialized bay located at the mouth of the Cowichan River near Duncan, British Columbia on Vancouver Island (48° 44.975' N, 123° 35.922' W) (Figure 3.1). Cowichan Bay has a high level of anthropogenic noise that originates from recreational boats entering and leaving two marinas, and from commercial activity (*e.g.,* tugboats and bulk carriers). Cowichan Bay also has a sawmill that stores log booms in the Cowichan estuary before being brought up to the mill to be processed.

Argonaut Wharf is located at the mouth of the Campbell River, in Campbell River, British Columbia and extends ~60 m into Discovery Passage, on the east coast of Vancouver

Island (50° 2.543’ N, 125° 14.834’ W). Argonaut Wharf also is in an area of high anthropogenic activity including a ferry that runs every 30 minutes to and from nearby Quadra Island, commercial vessels (*e.g.,* tugboats, cruise ships) transiting through, and recreational and fishing vessels entering and exiting a marina located 1 km away. Unlike Cowichan Bay, there is no sawmill near Argonaut Wharf.

3.3.2 Recordings

A SoundTrap (300 STD, Ocean Instruments, New Zealand) internally recording hydrophone system was deployed from Aug 15, 2019 to Oct 31, 2019 in Cowichan Bay, then redeployed from Aug 11, 2020 to Oct 22, 2020 in the same location (Figure 3.1B). The SoundTrap hydrophone was set to a sampling rate of 48 kHz at 16 bits and recorded for 15 minutes every hour. The SoundTrap was deployed in Cowichan Bay following the methods used in Murchy et al. (2022), on a polysteel line 1 m off the bottom (45 m water depth). Retrieval of the hydrophone was done using an acoustic release.

In 2021, the location was changed to Argonaut Wharf and deployed from Aug 13, 2021 to Sep 27, 2021 (Figure 3.1A). The SoundTrap was set to the same sampling rate (48 kHz) but recorded continuously throughout the deployment. The deployment method was also modified, the SoundTrap was secured to an underwater piling using cable ties, at a depth of 5 m off bottom (10 m water depth). Calibration of the SoundTraps was conducted by the manufacturer at 250 Hz with a source level of 120 dB re 1 µPa (B & K 2236 pistonphone).

3.3.3 Analysis

Data were stored as compressed sud files (.SUD) on the internal memory of individual SoundTraps until retrieval at the end of each deployment. All following data analysis was done using code written in Python (version 3.7). Sound Pressure Levels were examined in a time series for the full broadband frequency range of the recorder (20-24000 Hz). The soundscape at both locations was described using 5-minute averaged data.

Automatic Identification System (AIS) data from equipped vessels in a 5 km radius around Cowichan Bay was obtained for August through October of 2019 and 2020 in 5-minute bins, and 1-minute bins for a 10 km radius around Argonaut Wharf for August and September 2021. AIS data obtained were accurate to ± 1 m for the vessel's GPS location. All tugboat data found within the AIS data set were extracted and tracks were generated in ArcMap 10.8 (ESRI, California, USA). Distance between each tugboat's GPS location and the SoundTrap location were generated using the NEAR function to obtain the closest point of approach for each tugboat, and only tugs that were within 500 m of the SoundTraps were used in the analysis. Additionally, for the Argonaut Wharf dataset, tugboats needed to be within 500 m for at least 2 minutes (half of the 5-minute bins used for SPL calculations) to be included.

SPL measurements were binned into two equal time periods (with and without a tugboat present). Depending on the length of the tugboats' presence on a given day, the 'no tugboat' time period was selected to match. Power spectral density (PSD) plots were also generated for both time periods for the entire frequency range and compared. Sound pressure measurements between time points with and without tugboats were normally distributed, so t-tests were used for analysis, and were performed in RStudio version 4.1.2.

3.4 Results

3.4.1 Tugboat logging activities (Cowichan Bay)

Log transport activities did not occur during 2019 due to a strike at the Western Forest Mill located in Cowichan Bay, but re-started once the strike ended in early 2020. During August and September of 2020, six tugboats pulling log booms were detected within Cowichan Bay using AIS, however, only four times did the tugboat get within 500 meters of the hydrophone while it was recording.

Tugboats pulling logs near the estuary in Cowichan Bay significantly changed the soundscape for the full broadband range collected (20-24,000 Hz; Figure 3.2A). Median (5%, 95% CI) sound pressure levels near the estuary were 97.8 dB re 1 μPa (91.3; 106.4 dB re 1 μPa) when no tugboats were present. Once a tugboat with logs was within 500 m of the hydrophone, median sound pressure levels were significantly elevated to 107.6 dB re 1 μPa (98.0; 138.3 dB re 1 μPa) (t-test, $p < 0.0001$, df = 60, t = -7.3).

Tugboats pulling log booms into Cowichan Bay substantially changed the frequency composition near the estuary across the entire frequency range collected (10-24,000 Hz), at all percentiles (Figure 3.3A-D). Median increase in sound pressure level at different frequencies varied between 0.5 dB re 1μPa2/Hz (11 Hz) and 17.2 dB re 1μPa2/Hz (2216 Hz) when a log transport tugboat was present. However, the largest change was observed at 47 Hz with a 43.6 dB re 1μPa2/Hz increase in the 95th percentile.

3.4.2 Other tugboat activities (Argonaut Wharf)

Argonaut Wharf had more tugboat activities compared to Cowichan Bay, with 15 tugboats detected within 500 m of the SoundTrap hydrophone in 2021. However, only five tugs were detected within 500 m for longer than 2 minutes, and only a single tugboat was considered towing, as obtained from the AIS data (September 16). The other four tugboats could not be confirmed to be towing, but it also could not be determined that they were not.

Sound pressure levels (20-24,000 Hz) were higher in Argonaut Wharf compared to Cowichan Bay, but tugboats still had a significant impact on the underwater soundscape (Figure

3.2B). Median (5%, 95% CI) sound pressure levels were 122.7 dB re 1 μPa (91.3; 106.4 dB re 1 μPa) when no tugboats were present. Once a tugboat was within 500 m of the hydrophone median sound pressure levels were significantly elevated to 128.6 dB re 1 μPa (98.0; 138.3 dB re 1 μPa) (t-test, $p = 0.02$, $df = 33$, $t = -2.3$).

Tugboats present in Argonaut Wharf only slightly altered the frequency composition at any of the percentiles examined, with only approximately 40% of frequencies elevated (Figure 3.4A-D). Median increase in sound pressure level at different frequencies varied between 0.0002 dB re 1μPa2/Hz (12004 Hz) and 6.1 dB re 1μPa2/Hz (1155 Hz) with a tugboat present. However, the largest change was observed at 7 Hz with a 12.6 dB re 1μPa2/Hz increase in the 95th percentile.

3.5 Discussion

The presence of tugboats had a significant impact on the underwater soundscape in both locations considered. In Cowichan Bay, tugboats used for log transport activities caused short term changes to the soundscape by elevating the sound pressure level near the estuary, across all frequencies collected. Similarly in Argonaut Wharf, sound pressure levels were increased, but frequency composition was similar between time points. The differences observed between the two locations indicates that the impact of tugboats on the underwater soundscape is context specific and varies between locations.

Log transport tugboats present in Cowichan Bay increased the soundscape noise levels near the estuary adding an additional impact to critical habitat already degraded by log storage activities. Currently in British Columbia, timber is stored as rafts in estuaries prior to processing at mills, which can lead to changes to benthic communities through bark being deposited on the ocean floor, and compaction of the sediment (Toews and Brownlee, 1981). Bark deposition has also been linked to reduced zooplankton abundance and dissolved oxygen (Power, 1987). Additionally, the lower oxygen levels near log storage areas diminished the number of juvenile salmon in those areas, potentially displacing them (Levy et al., 1990). Results from this chapter demonstrate an additional stressor, underwater noise, that requires additional research into its impact to the marine animals that live in the estuary.

Argonaut Wharf had higher sound pressure levels compared to Cowichan Bay when no tugboats were present. This difference is likely due to the high presence of anthropogenic activities in the Strait of Georgia (Erbe et al., 2012). One noise source not found in Cowichan Bay that was present in Argonaut Wharf was passenger ferries. A car and passenger ferry runs from Campbell River to Quadra Island twice an hour transiting between 1.7-2.0 km away from the hydrophone location. When examining the contribution (*i.e.*, number of vessels, time present and source level) of ferries to the underwater soundscape in the Strait of Georgia, passenger ferries account for a high relative noise contribution (approximately 70-88%), but the exact noise contribution varies depending on the time of year (MacGillivray et al., 2016). In comparison, tugboats in the Strait of Georgia account for roughly 7-22% of the relative noise contribution to the area (MacGillivray et al., 2016).

Significant impacts to the underwater soundscape were observed in the study presented in this chapter, but the study was not without its limitations. The study was only conducted over short time periods between August and October. During these time windows Chinook salmon (*Oncorhynchus tshawytscha*) are returning to spawn in both locations. Cowichan Bay has a First Nation subsistence fishery while Argonaut Wharf has an open recreational fishery directly in front of the deployed hydrophone, resulting in higher amounts of recreational vessel activities in this area which could be minimizing the observed impacts. Additionally, due to the local geography of the two sites, the data collected for this chapter could only be used to examine the impact of tugboats on underwater soundscape on limited spatial scales (500 m). Larger scale spatial impacts could not be evaluated. However, estimates of long-distance impacts can be generated using the sonar equation to calculate where the noise produced by tugboats would equal background noise levels. Source levels of tugboats collected in Cowichan Bay and at Argonaut Wharf were back-calculated from the median sound pressure levels when tugboats were within 500 m and were 137 dB re 1 µPa @ 1m and 155 dB re 1 µPa @ 1m, respectively. In Cowichan Bay, impacts of tugboat noise were estimated to be above median ambient sound levels (97.8 dB re 1 µPa) until approximately 10 km away from the hydrophone. Argonaut Wharf had higher median ambient sound levels (122.7 dB re 1 µPa), which resulted in a shorter distance (2 km) at which the tugboat noise was above ambient noise levels. Overall, tugboat noise could be altering the underwater soundscape between 2-10 km away from the vessel. Future work should evaluate the impact of tugboats on the underwater soundscape on seasonal scales and a range of spatial scales to account for seasonal variation in vessel activities and oceanographic conditions.

Both locations used in this chapter represent critical habitat for many marine species, but the influence of the noise produced by tugboats on these species has not been evaluated. Resident killer whales (*Orcinus orca*) display avoidance (horizontal and vertical: Williams et al., 2009, 2002) and change foraging behaviours (Lusseau et al., 2009) around vessels. Similarly, Pacific herring (*Clupea pallasii*) and juvenile salmon (*Oncorhynchus* spp.) have behavioural responses to elevated sound pressure levels produced from vessels (van der Knaap et al., 2022). Recent meta-analyses have also documented widespread impact of shipping noise on fish and invertebrates (Cox et al., 2018; Murchy et al., 2020), indicating broad implications for elevated noise levels due to shipping activities. Future research should continue to evaluate the impacts of underwater noise on marine species, and tugboats need to be represented in these studies.

3.6 Conclusions

Large commercial vessels have been documented to increase underwater noise levels, with mitigation methods already being implemented (MacGillivray et al., 2019; Williams et al., 2019). In British Columbia, tugboats represent a significant contributor to local underwater noise, with the highest total vessel hours for all AIS vessel categories (MacGillivray et al., 2016). The work presented in this chapter demonstrates that tugboats significantly elevate ambient underwater sound levels when present, representing an understudied contributor to the underwater soundscape. Future work should continue to examine the impact of smaller commercial vessels, like tugboats, to the underwater soundscape and marine species to help develop mitigation measures to reduce their impacts.

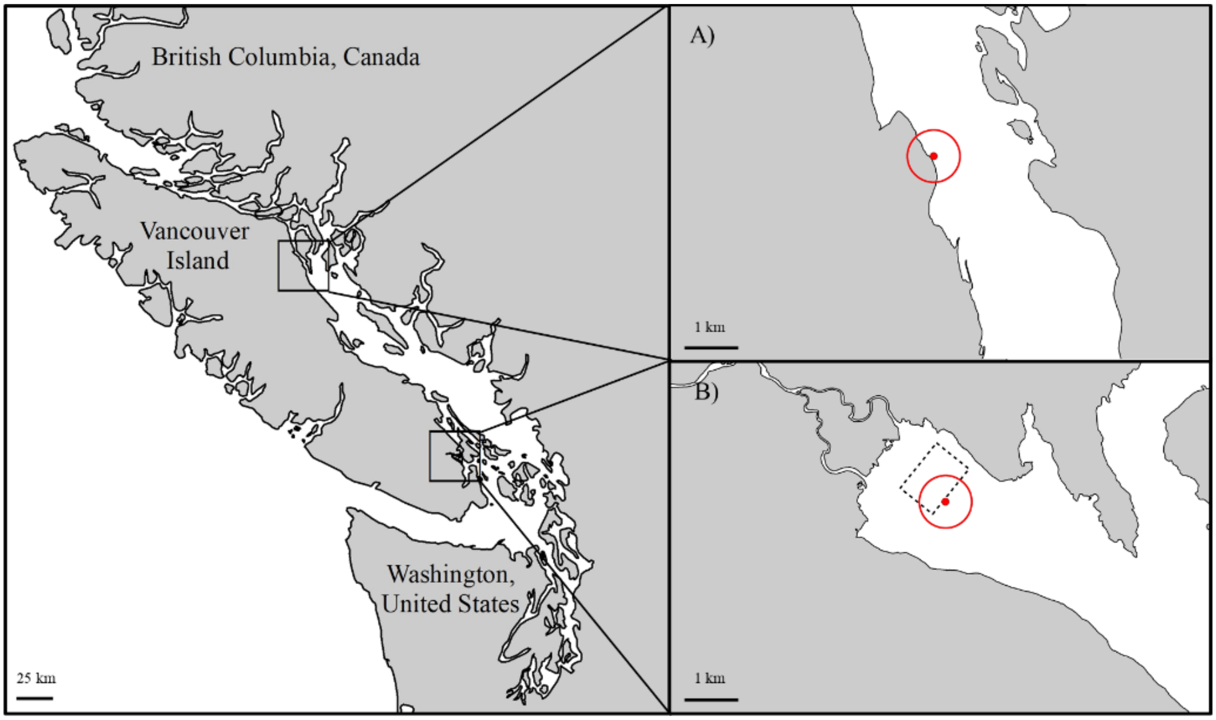

Figure 3. 1. Map of Vancouver Island, British Columbia. A) Argonaut Wharf in Campbell River, BC. Red dot shows SoundTrap hydrophone mooring location. B) Cowichan Bay, BC. Red dot shows SoundTrap hydrophone mooring location and black dashed box denotes the log boom sorting area in the Cowichan River estuary. Map shapefiles were obtained from the Natural Earth and BC Provincial data catalog.

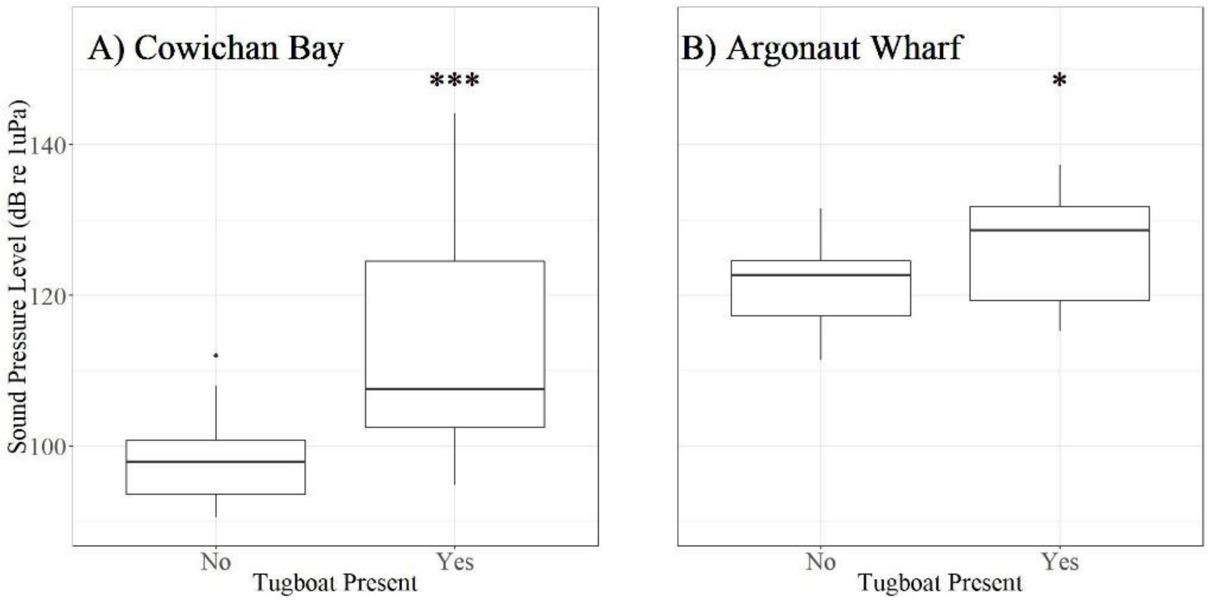

Figure 3. 2. Boxplots of sound pressure levels (broadband: 20-24,000 Hz) in A) Cowichan Bay and B) Argonaut Wharf without and with a tugboat within 500 m of hydrophone. Asterisks (***) indicate significantly different mean sound pressure levels at $p < 0.0001$ and a single asterisk (*) indicates significance at $p = 0.02$. Significance was tested using paired t tests.

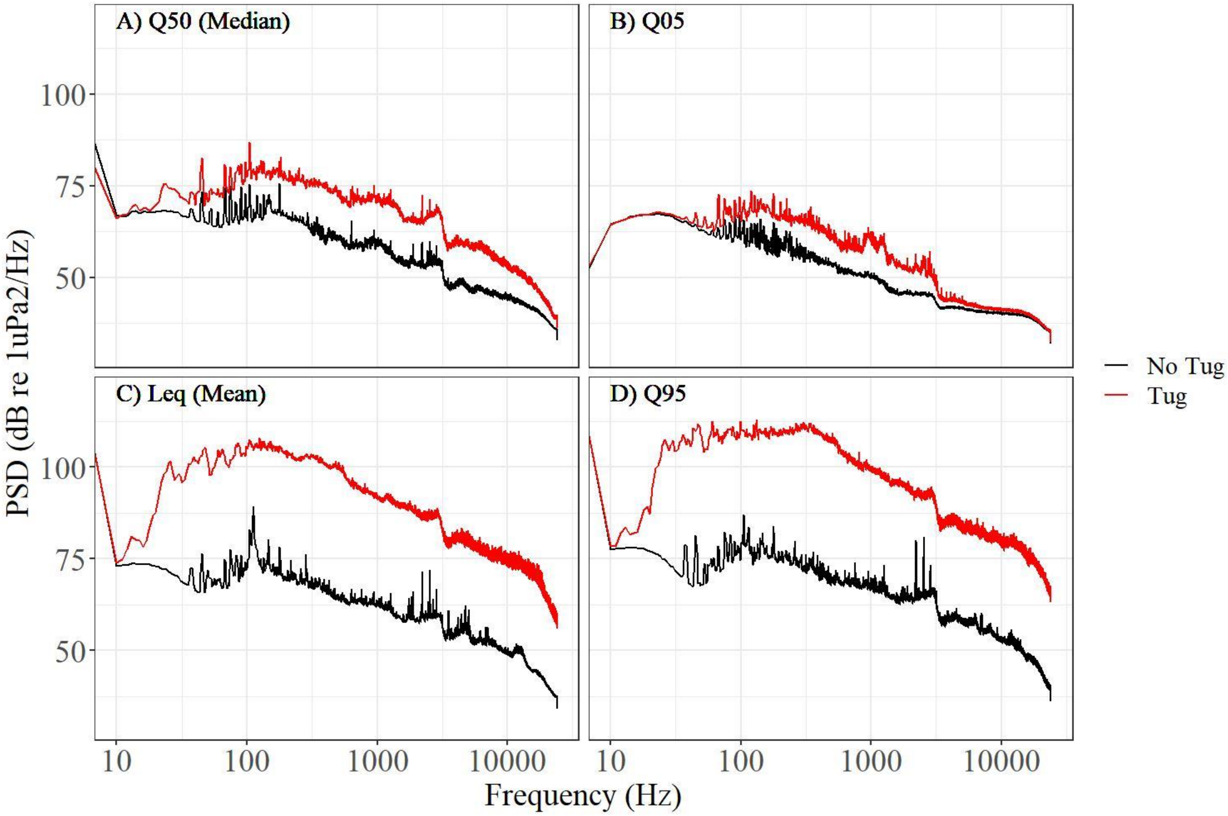

Figure 3. 3. Power Spectral Density (PSD) plots of logging tugboats in Cowichan Bay (A) median values (50th percentile), (B) 5th percentile, (C) mean values (Leq), and (D) 95th percentile with no tugboats (black) and while tugboats were moving logs into the estuary (red).

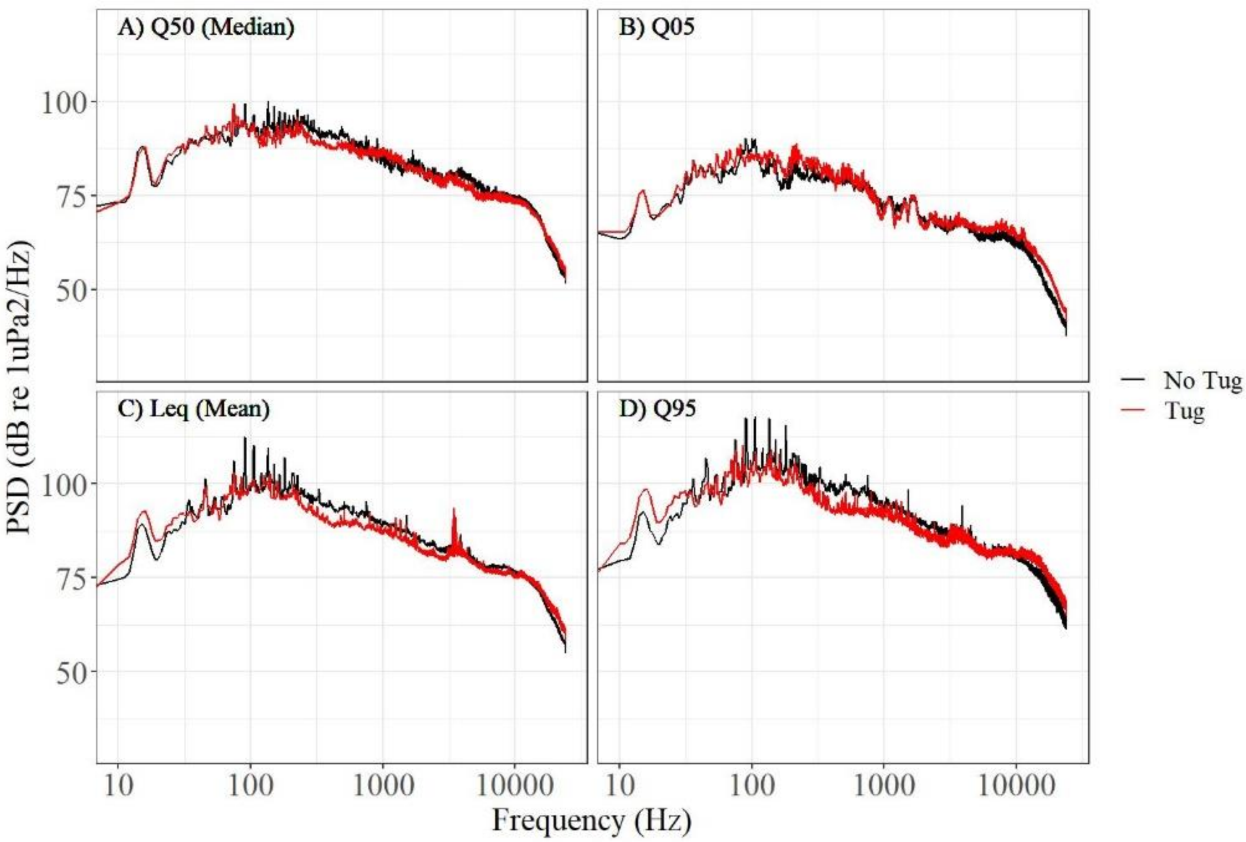

Figure 3. 4. Power Spectral Density (PSD) plots of tugboats in Argonaut Wharf (A) median values (50th percentile), (B) 5th percentile, (C) mean values (Leq), and (D) 95th percentile with no tugboats (black) and with tugboats within 500 m of hydrophone (red).

Chapter 4 - Preliminary examination of sounds produced by Pacific salmon (*Oncorhynchus* spp.) during their fall spawning migration

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4.1 Abstract

A preliminary description of sounds produced by three species of Pacific salmon was conducted to address the lack of quantified call characteristics in previous studies. Wild Chinook salmon (*Oncorhynchus tshawytscha*), pink salmon (*O. gorbuscha*) and coho salmon (*O. kisutch*) were diverted from a natural spawning migration in the Big Qualicum River located on Vancouver Island, British Columbia, Canada and held in the adjacent hatchery during the 2017 fall migration. Underwater sounds were opportunistically recorded continuously over four weeks in holding raceways containing Chinook only, coho only, or mixed pink and Chinook salmon, and examined for sounds. All groups produced sounds in three categories based on mechanism: hydrodynamic (surface splash), air movement (miscellaneous and 7 named types), and unknown mechanism (pulse). Pulse, gill-bubble FRT air movement sounds, and miscellaneous air movement sounds occurred in all groups and differences in some characteristics of sounds were found between the species groups. Additionally, even though pink salmon were not recorded separately, data suggest they produce a very fast repetitive tick (vFRT) air movement sound more often compared to Chinook salmon. Our results represent the first detailed description of the types and characteristics of sounds produced by wild Pacific salmon.

4.2 Introduction

To date 1,185 different species of fish have been documented to produce sounds, across 133 different families (Looby et al., 2023, 2022). Among these, 15 species of salmonids have been documented to produce sounds (Looby et al., 2022; Murchy et al., 2023), although most of the information on sounds produced is descriptive with limited information on characteristics. Salmonid sound production can be broken down into four categories: air movement (Rountree et al., 2018), percussion (Neproshin and Kulikova, 1975), substrate sounds (Stober, 1969) and hydrodynamic sounds (Neproshin and Kulikova, 1975; Rountree et al., 2018). Air movement sounds are the most common (Rountree et al., 2018) and originate from gas release through the anus or gills but also can be produced by internal movements of gas between the gas bladder and the pneumatic duct (Neproshin and Kulikova, 1975). For example, fast repetitive ticks (FRTs) similar to those observed in herring (Wilson et al., 2004) are also produced by Arctic char (*Salvelinus alpinus alpinus)* (duration: 98-107 ms; frequency range: 690-760 Hz; Bolgan et al., 2016). Rountree et al. (2018) found similar types of sounds produced by Atlantic salmon (*Salmo salar)*, brook trout (*Salvelinus fontinalis)*, brown trout (*Salmo trutta)* and rainbow trout (*Oncorhynchus mykiss)*. Brook, brown and rainbow trout all produce sounds like FRTs but shorter in duration known as very fast repetitive ticks (vFRT), but this sound was not observed in Atlantic salmon (Rountree et al., 2018). However, Atlantic salmon and rainbow trout both produce a gurgle sound at peak frequencies of 748 Hz and 2,409 Hz, respectively (Rountree et al., 2018). When similar sounds were produced by different species (*e.g.*, vFRTs), frequency and duration characteristics were slightly different and sounds could be separated out by species in a multivariate analysis (Rountree et al., 2018). Other air movement sounds are species specific. Brook trout produce a snitch (peak frequency: 4,617 Hz) and brown trout produce a chirp (peak frequency: 4,760 Hz), while Atlantic salmon produce a "moan" sound (peak frequency: 943 Hz; Rountree et al., 2018). Hydrodynamic sounds, which are associated with air movement sounds due to gulping air at the surface, can also be species specific within limited context. For example, some species (*e.g*., brook trout) are quiet at the surface while others produce loud splashes or jumping (*e.g.*, rainbow trout; Rountree et al., 2018).

Some sounds produced by salmonids appear to be potentially linked with spawning behaviour (Johnson et al., 2018; Stober, 1969). Substrate sounds (700 Hz – 22 kHz; inferred from published spectrograms) from redd building have been documented in Arctic char (Bolgan et al., 2016) and cutthroat trout (*Oncorhynchus clarkii*; Stober, 1969), and are potentially important for gamete release (Moore and Waring, 1999). Additionally, Johnson et al. (2018) documented that growl sounds (peak frequencies: 20 and 50 Hz) were produced by lake trout (*Salvelinus namaycush*) only on the spawning reef, indicating vocalizations associated with spawning behaviours. Percussive and stridulation noises from jaw movements, could also be courtship sounds (Dubois and Dziedzic, 1989; Johnson et al., 2018). Lake trout produce snaps (frequency range: 20-170 Hz) from jaw movement or nudging behaviours (Johnson et al., 2018), while European whitefish (*Coregonus lavaretus*) produce scraping sounds (frequency range: 100-300 Hz) during close contact between males and females (Dubois and Dziedzic, 1989).

Pacific salmon (*Oncorhynchus* spp.) are important species in the Northeast Pacific Ocean, with numbers declining in recent years (Beamish et al., 1999). Past research on sound production in Pacific salmon comes mainly from the 1970's (Neproshin, 1972; Neproshin and Kulikova, 1975) and lacked detailed descriptions of sound characteristics. Kuznetsov (2009) was the first to examine some sound characteristics in pink (*Oncorhynchus gorbuscha*) and chum salmon (*Oncorhynchus keta*), but the full range of sounds produced by Pacific salmon species has yet to be evaluated. The aim of this paper is to conduct a preliminary qualitative examination of sounds produced by three species of wild Pacific salmon during their fall spawning migration (mid-August to mid-November) and start characterizing parameters (duration, frequency range, peak frequency, etc.) of the different sound types found.

4.3 Methods

4.3.1 Study location

Big Qualicum Hatchery is located in Qualicum Beach, British Columbia (49° 20.894' N, 124° 26.570' W), at river kilometer (km) 1 on the Big Qualicum River. The hatchery consists of three flow-through (0.5L/second) concrete raceways (61m x 4.5m x 1.2m) that contain river water (Figure 4.1). Various species of Pacific salmon spawn in Big Qualicum River and as the wild salmon move upriver, they are diverted into a constructed channel that places them in the first raceway (Raceway 3: Figure 4.1) of the hatchery. Since different species might come upriver at similar times, once in the first raceway the hatchery staff sorts the salmon by species into different raceways for holding (Raceway 1 and 2: Figure 4.1). Stocking densities in each raceway were visually assessed to be no more than 18-22 individuals/m^3 by hatchery staff.

4.3.2 Sound recordings

A SoundTrap STD300 (Ocean Instruments, New Zealand, system gain: -171.5 to -175.4 dB re FS/µPa, flat frequency response from 10 Hz to 72 kHz, self-noise < 34 dB re 1 µPa above 2 kHz) underwater acoustic recorder was deployed in each raceway (raceway 1 and 2, Figure 4.1). Each SoundTrap was placed on the bottom (water depth: 1.2 m) approximately 30.5 m from the south side and 2 m from west side of each raceway (Figure 4.1). Due to the two-week autonomy of the SoundTrap recorders, data were collected in two deployments. For the first deployment (September 18 to October 03, 2017), the recorders were set to record continuously from 10 Hz to 48 kHz (*i.e.*, sampling rate: 96 kHz) with a 16-bit depth. For the second deployment (October 03 to October 17, 2017), the sampling rate was adjusted to 48 kHz (10 Hz to 24 kHz) to reduce data storage (Table 4.1). For both deployments, the SoundTrap were also configured to record temperature every minute. During September and October of 2017, 1341.8 hours (55.9 days) of audio files were collected with three different species of wild Pacific salmon present at various times (pink salmon, *Oncorhynchus gorbuscha,* Chinook salmon, *Oncorhynchus tshawytscha* and coho salmon, *Oncorhynchus kisutch*). All salmon present in the hatchery were maturing adult fish, length measurements were not recorded but each species was likely similar sized, with size differences primarily between species. Chinook salmon and coho salmon were present at different times in mono-species groupings, however, pink salmon were always mixed with Chinook salmon when present. Temperature was relatively consistent throughout the study with both raceways remaining around 9.5° C ± 1° C.

4.3.3 Sound documentation

Prior to processing, all audio files collected during the first deployment, were downsampled to a 48 kHz using Sox-o-matic (version 1.1.10; Cornell University, Ithaca New York, USA) to match audio files collected during the second deployment. All processing was conducted in Raven Pro acoustic software (Raven Pro, version 1.5). Sound files were processed in two sets, resulting in 28 days analyzed for sound production; 1) a 24 h block of time was selected from hatchery notes and was examined in its entirety for the single species groups (Chinook and coho) to maximize the likelihood of detecting all sound types; 2) a subset of 1-min clips from every hour and every other day were selected from the full time series for each species group to examine diel trends in occurrence (Table 4.1). In both cases, spectrograms and waveforms of sounds were visually examined by an analyst (KM) to identify potential sounds (Spectrograms with 1,024-sample FFT, Hanning window, 50% overlap), and confirmed by a second analyst (RR).

All sounds were categorized into three broad types based on production mechanism: hydrodynamic sounds (surface event associated air gulping), air movement sounds (*sensu* Rountree et al., 2018), and unknown mechanism sounds. Additionally, air movement sounds were broken down into specific sound types (*e.g.*, vFRT) when possible following names used in previous salmonid studies (Bolgan et al., 2016; Rountree et al., 2018). Acoustic measurements were made in Raven for each sound including: its low frequency, 1st quartile frequency (25%), peak frequency, median frequency (50%), 3rd quartile frequency (75%), high frequency, bandwidth (high-low), 90% bandwidth, duration, and 90% duration (Charif et al., 2010). Additionally, multiple sounds that occurred in close proximity (maximum 4 seconds between sounds) and appeared to be part of a single sound sequence, often following a surface event sound, were grouped and labelled as a "Sound Series" (*sensu* Rountree et al., 2018). The number of individual sounds, number of unique sound types, total duration (start of first sound to end of last sound), duration of surface event, latency (time from start of surface event to start of first salmon sound), and duration of each salmon sound were measured for sound series that included a surface event.

4.3.4 Statistical Analysis

Descriptive statistics (mean, SE, minimum and maximum) were tabulated for each sound variable measurement (*e.g.*, peak frequency) for each sound type, as well as sound series, within each species group. Diel patterns in sound production were examined by counting the number of occurrences of each sound type by species for each hour of the day in which each group was present and analyzed (Chinook: $n_1 = 8$; coho: $n_1 = 2$; mixed: $n_1 = 7$; Table 4.1; n_1 = number of days with 24-hour subsamples). Mean and SE were then calculated for each species/sound type combination by hour of the day. Each species was additionally pooled for all types of sounds to examine overall diel trends in sound production. Sound characteristics were compared between the coho only and Chinook only groups, as well as between the Chinook only and mixed Chinook and pink salmon group, by plotting the mean ± SE of each acoustic metric. All analyses were conducted in RStudio Version 4.1.2.

4.4 Results

Various types of sounds were found in all three groupings, indicating sound production in Chinook and coho salmon and potential sound production in pink salmon. A total of 28.3 hours of audio files were analyzed when only Chinook salmon were present in the raceways (out of 535.2 hours collected, Table 4.1), and resulted in a total of 319 sounds identified, and 44 series events (Table 4.2; Figure 4.2). Coho salmon were recorded from one raceway over a 109 hour period (Table 4.1), in which 147 sounds, and 18 series events were detected in the 24.4 h analyzed (Tables 4.2; Figure 4.3). Pink salmon and Chinook salmon were mixed in both raceways for the first deployment (Sept 18- Oct 02), resulting in 696.9 hours collected but only 5.6 hours were analyzed (Table 4.1) resulting in 162 sounds, and 22 series events (Tables 4.2; Figure 4.4).

4.4.1 Surface events

Surface events created from hydrodynamic sounds associated with surface movement and air gulping were detected in all three groupings (Table 4.2). For Chinook salmon, a total of 44 surface series events were collected (Figure 4.2A). Total sound series duration was 2.23 s ± 0.18

s, with a surface event duration of 0.92 s ± 0.09 s, latency of 1.43 s ± 0.13 s and a fish sound duration of 0.80 s ± 0.14 s. The total number of fish sounds occurring in a series varied from one to four sounds and were composed of one to two unique sound types.

Mm. 1: Chinook salmon surface event followed by miscellaneous air movement sound. This is a file of type .wav (302 KB).

Surface series events produced by coho salmon, occurred 18 times in the dataset (Figure 4.3A). Total sound series duration was 1.77 s ± 0.16 s, with a surface event duration of 0.55 s ± 0.07 s, latency of 1.00 s ± 0.10 s and a fish sound duration of 0.77 s ± 0.18 s. The number of fish sounds occurring in a series varied from one to four sounds and were composed of a mean of 1.28 ± 0.18 unique sound types (air movement, pulse, etc.) observed.

Mm. 2: Coho salmon surface event followed by miscellaneous air movement sound. This is a file of type .wav (280 KB).

In the mixed school surface sound series occurred 22 times (Figure 4.4A). Total sound series duration was 1.76 s ± 0.14 s, with a surface event duration of 0.79 s ± 0.10 s, latency of 1.18 s ± 0.13 s and a fish sound duration of 0.57 s ± 0.14 s. Number of fish sounds occurring in a series varied from one to four sounds and varied in sound types observed with one or two unique sound types observed in a single series.

Mm. 3: Mixed Chinook and pink salmon surface event followed by miscellaneous air movement sound. This is a file of type .wav (392 KB).

4.4.2 Air movement sounds

Air movement sounds that are assumed to originate from gas release out of the swim bladder or from internal movements within the gas bladder were also observed in all three groups (Table 4.3). Seven different types of air movement sounds were produced by Chinook salmon

(Table 4.3; Figure B4.1). One common class of air movement sound was labeled a “gill-bubble FRT” due to its qualitative similarity to a sound reported to be produced by bubble release through the gills in brook trout (Rountree et al., 2018). Gill-bubble FRTs occurred in 16.6% (relative occurrence) of the sounds annotated, and had a peak frequency of 630 Hz ± 45 Hz and a duration of 0.78 s ± 0.06 s (Figure 4.2C). Additional air movement types were moan (Figure 4.2B) and vFRT sounds (Figure 4.2F) that were found at a similar level of occurrence (4.5% and 5.4% relative occurrence, respectively). Moan sounds had a peak frequency of 405 Hz ± 42 Hz and a duration of 0.39 s ± 0.09 s, while vFRTs are higher frequency (peak frequency: 2029 Hz ± 361 Hz) and longer duration (0.51 s ± 0.26 s). The final three sound types, gurgle, buzz, and growl occurred less often (< 5.0%; relative occurrence)(Table 4.3: Figure B4.1). The remaining air movement sounds that could not be classified from previous naming systems were grouped into miscellaneous air movement sounds. Miscellaneous air movement sounds were also common (30.7% relative occurrence) and were characterized by a peak frequency of 829 ± 79 Hz and a duration of 0.30s ± 0.03 s (Figure 4.2D).

Mm. 4: Chinook salmon series including a gill-bubble FRT and moan sound. This is a file of type .wav (323 KB).

Mm. 5: Chinook salmon gill-bubble FRT sound. This is a file of type .wav (236 KB).

Mm. 6: Chinook salmon miscellaneous air movement sound. This is a file of type .wav (226 KB).

Mm. 7: Chinook salmon vFRT sound. This is a file of type .wav (207 KB).

Coho salmon produced six different types of air movement sounds (Table 4.3). The most common (25.9% relative occurrence) classified air movement sound documented for coho salmon was the gurgle sound (peak frequency: 622 Hz ± 36 Hz; duration: 0.40 s ± 0.08 s) (Figure

4.3F). The gill-bubble FRT (peak frequency: 678 Hz ± 70 Hz; duration: 1.03 s ± 0.13 s) sounds were also common for coho (~10.0% relative occurrence each) (Figure 4.3C). The final three classified air movement sound types included moan, vFRT, and whistle, but these sounds were relatively rare with the moan only occurring three times and vFRT and whistle only occurring once each (Figure 4.3B; Figure B4.2). The remaining air movement type sounds that could not be classified were grouped into miscellaneous air movement sounds which had a peak frequency of 894 Hz ± 112 Hz and duration: 0.29 s ± 0.07 s (Figure 4.3E).

Mm. 8: Coho salmon series including a gill-bubble FRT, miscellaneous air movement sounds, pulse sound and a whistle sound. This is a file of type .wav (302 KB).

Mm. 9: Coho salmon gill-bubble FRT sound. This is a file of type .wav (268 KB).

Mm. 10: Coho salmon miscellaneous air movement sound. This is a file of type .wav (218 KB).

Mm. 11: Coho salmon gurgle sound. This is a file of type .wav (109 KB).

Nine different air movement sound types were identified when pink and Chinook salmon were mixed in the same raceway (Table 4.3). The most common type was the vFRT sound which occurred at 21.4% of the sounds observed (Figure 4.4E) and had a peak frequency of 3655 Hz ± 32 Hz and a duration of 0.12 s ± 0.10 s. Other sounds that were documented included a gill-bubble FRT sound (peak frequency: 797 Hz ± 104 Hz; duration: 0.43 s ± 0.09 s) (Figure 4.4F) which occurred less than 10.0% relative occurrence. Rarer sound types included: buzz, gurgle, raspy, and FRT occurred at a lower relative frequency of occurrence (< 2.0%), while the growl and moan sounds, occurred only once each (Figure B4.3). Similarly, to Chinook only and coho salmon the remaining sounds were grouped together. Miscellaneous air movement sounds were common (45.2% relative occurrence) in the mixed school, and had a peak frequency of 1135 Hz ± 64 Hz and a duration of 0.15 s ± 0.20 s (Figure 4.4C).

Mm. 12: Mixed Chinook and pink salmon series including miscellaneous air movement sound and a pulse. This is a file of type .wav (314 KB).

Mm. 13: Mixed Chinook and pink salmon gill-bubble FRT sound. This is a file of type .wav (507 KB).

Mm. 14: Mixed Chinook and pink salmon miscellaneous air movement sound. This is a file of type .wav (405 KB).

Mm. 15: Mixed Chinook and pink salmon vFRT sound. This is a file of type .wav (402 KB).

4.4.3 Unknown Mechanism sounds

A short duration sound classified as a pulse sound was identified in all three groups, but the mechanism for this sound is unknown. Pulse sounds were the most common type observed in Chinook salmon accounting for about 39.6% (relative occurrence). Chinook pulse sounds were short duration (0.06 s ± 0.003 s) sounds with a high peak frequency (1435 Hz ± 117 Hz) (Figure 4.2E). Pulse sounds were also the most common sound produced by coho salmon with a relative occurrence of 50.3% and a peak frequency of 1204 Hz ± 78 Hz and a duration of 0.07 s ± 0.01 s (Figure 4.3D). Pulse sounds were also observed in the mixed school but at a lower occurrence (19.6% relative occurrence), with a peak frequency of 2278 Hz ± 285 Hz and a duration of 0.05 s ± 0.005 s (Figure 4.4D).

Mm. 16: Chinook salmon pulse sound. This is a file of type .wav (196 KB).

Mm. 17: Coho salmon pulse sound. This is a file of type .wav (199 KB).

Mm. 18: Mixed Chinook and pink salmon pulse sound. This is a file of type .wav (392 KB).

4.4.4 Diel patterns

When all sound types are pooled, all groups (Chinook, coho, and pink and Chinook mixed) produced sounds at a consistent rate throughout the day, however, some individual sound types exhibited diel patterns. Chinook salmon did not appear to have any pattern in sounds produced for either pooled sound types, or any individual sound type, producing sounds consistently throughout the day (Figure B4.4 and B4.5). Coho salmon had diel patterns in miscellaneous air movement sounds, where sound production was elevated between 12:00 and 15:00, peaking at 14:00 (Figure 4.5A), but no other sound types had a diel pattern (Figure B4.5). The pink/Chinook mixed group had a strong, diel pattern only for the vFRT sounds with more produced between 11:00 and 17:00 (Figure 4.5B).

4.4.5 Comparisons between species

All three species groupings had some of the same sound types in each of the different sound categories (surface, air movement and unknown), however, each species grouping had unique sound types and different rates of occurrence of similar sounds (Table 4.3). Since not all sound types were observed in each grouping, comparisons of acoustic characteristics were conducted only on common sound types.

Comparison of surface series events between Chinook salmon and coho salmon showed that most variables were similar between the two groups (series duration, latency, number of sounds, sound durations) but surface event duration was shorter for coho salmon compared to Chinook salmon (Table 2). A lower number of overall sound types (Table 4.3) were also documented for coho salmon (seven) compared to Chinook salmon (eight), but five sound types were produced by both Chinook salmon and coho salmon, including miscellaneous air movement, gill-bubble FRT, gurgle, moan and pulse sounds. Pulse sounds, which were the most common sound type for both Chinook and coho salmon, had some higher frequency metrics (peak, median, 3rd quartile, high), a wider frequency bandwidth (full and 90%; Figure B4.6B) and shorter duration (full and 90%) (Figure B4.7) in Chinook salmon compared to coho. The second most common sound produced by coho salmon were the gurgle sound but were uncommon in Chinook salmon. Gurgle sounds did not appear to be different for most metrics but did appear to have a higher high frequency and wider full bandwidth in Chinook compared to coho salmon (Figure B4.8B and B4.7). Miscellaneous air movement sounds were more prevalent in Chinook salmon compared to coho salmon and had a higher high frequency creating a wider bandwidth (90% and full) in Chinook salmon compared to coho salmon (Figure B4.6A). One sound found in similar frequency between Chinook and coho salmon was the gill-bubble FRT. Gill-bubble FRT sounds had a lower low frequency and 1st quartile frequency in Chinook compared to coho (Figure B4.8A). Additionally, Chinook salmon had a higher high frequency and a wider bandwidth (90% and full), while also having a shorter duration and 90% duration (Figure B4.7). Finally, the moan sounds were more common in Chinook than either coho or mixed chinook and pink salmon (Table 4.3).

When comparing surface series events produced between Chinook salmon and pink/Chinook mixed groups, all variables were similar between the two groups (surface duration, latency, number of sounds). All sound types identified in the Chinook only were also detected in the pink/Chinook mixed group, but the mixed school had two additional uncommon types detected (Raspy and FRT). Four of the common sound types (gurgle, moan, growl, and buzz) occurred less than 5.0% (relative occurrence) in both groups so were not compared, but the other four sound types were examined for comparisons (miscellaneous air movement, gill-bubble FRT, vFRT, and pulse sounds). Like the Chinook salmon only group, mixed schools of pink and Chinook salmon commonly produced miscellaneous air movement and pulse sounds, but at different frequencies of occurrence. Miscellaneous air movement sounds were produced more often in the mixed school compared to Chinook only. While the opposite occurred for pulse sounds, with pulse sounds occurring more often in Chinook only and less often in mixed groups. Both miscellaneous air movement and pulse sounds produced in the pink/Chinook mixed group had higher frequency metrics (1st quartile, peak, median, 3rd quartile, and high frequency), wider bandwidth (full and 90%) and shorter duration (full and 90%) compared to the only Chinook group (Figure B4.6 and B4.7). Similarly, gill-bubble FRT sounds also occurred less often in the mixed school compared to Chinook only and had a higher low frequency, 1st quartile frequency, peak frequency, median, and 3rd quartile frequency in the mixed group compared to Chinook only and also were shorter in duration (90% duration and full duration)(Figure B4.8A and B4.7). One sound type that occurred at a higher rate in the mixed group compared to the Chinook only was the vFRT sound. vFRT sounds produced in the mixed group also had higher low frequency, 1st quartile frequency, peak frequency, median, and 3rd quartile frequency, with a lower high frequency resulting in a shorter bandwidth (full) compared to Chinook only (Figure B4.9). Lastly, vFRT sounds produced by the mixed school were shorter duration (full) compared to Chinook only (Table 4.3; Figure B4.7).

4.5 Discussion

Wild Chinook salmon, coho salmon and mixed Chinook and pink salmon schools were all documented to produce sounds as adults returning to the river to spawn. Although, previous research has reported sound production by each of these species (Kuznetsov, 2009; Neproshin, 1972; Neproshin and Kulikova, 1975), detailed description of the sound types and characteristics are reported for the first time, herein. All three groups produced miscellaneous air movement, pulse, and gill-bubble FRT sounds consistently in the data analyzed, however, each group produced sounds at different rates of occurrence and had some unique sound types.

Our measurements indicated that sound parameters might vary between different species, similar to what has been observed in other salmonids (Rountree et al., 2018). Chinook salmon and coho salmon were documented to produce different types of sounds, have different frequencies of occurrence, and qualitative differences in sound metrics between the two species, indicating the potential for species specific sounds. Previous research has demonstrated species specific air movement sounds compared among four other species of salmonids using multivariate analysis (Rountree et al., 2018). The differences in frequencies between species could potentially be based on fish size as is seen in other species (Connaughton et al., 2000; Kuznetsov, 2009), since Chinook salmon are typically the largest of the Pacific salmon and pink salmon are the smallest (Beamish et al., 2018; Godwin and Krkosek, 2022; Radchenko et al., 2018; Riddell et al., 2018). Additionally, coho salmon had shorter mean durations of surface events (0.55s) compared to Chinook salmon (0.92 s), suggesting potential differences in surface behaviour (*i.e.*, air gulping) between Pacific species, as has been similarly reported for other salmonids (Rountree et al., 2018).

Even though pink salmon were never alone in the raceways, it is likely that the vFRT sound was primarily produced by pink salmon. The vFRT sound documented with the mixed school was produced at a higher occurrence of 21.4% relative occurrence compared to only 5.4% relative occurrence in the Chinook only group, despite a greater sampling effort for Chinook salmon. The vFRT sound documented in the mixed school was also at a higher frequency compared to the Chinook salmon only group for most frequency metrics (low, 1st quartile, peak, median, and 3rd quartile) and was shorter duration. Additionally, the difference in diel patterns of vFRT sounds was only observed in the mixed school and not when Chinook were alone in the raceways. Taken together, the differences between frequency metrics, duration, and temporal pattern suggests that the vFRT sounds produced by pink salmon differed substantially from that of Chinook, however future work should examine pink salmon in mono specific schools to confirm this conclusion.

No previous work has examined diel patterns in coho or Chinook salmon sound production. We found no evidence for a diel pattern for Chinook, but coho demonstrated one sound type (miscellaneous air movement sounds) with a possible diel pattern (Figure 4.5A. However, coho were only examined on two days, so more research would need to confirm if coho salmon do indeed have a diel pattern. In contrast, pink salmon have been previously documented to have a diel pattern in sound production, with more sounds produced between 22:00 and 02:00 (Kuznetsov, 2009). While Kuznetsov (2009) did not label the sounds they documented as vFRT sounds, their work sets a precedent for diel patterns in air movement sound production by pink salmon. The observed diel patterns in occurrence of the vFRT sounds in the mixed Chinook and pink salmon group (Figure 4.5B), is consistent with Kuznetsov's (2009) report of diel sound production behaviour in pink salmon.

Even though many sounds documented in this study were above the salmonid known hearing ranges (Amoser et al., 2004; Hawkins and Johnstone, 1978; Mann et al., 2007; Oxman et al., 2007), they are important components of the aquatic soundscape ecology (reviewed in Murchy et al., 2023). One example is the sounds associated with redd building. To build a redd, salmon must move sediment, generating noise (> 1 kHz: example in Murchy et al., 2023) above their documented hearing threshold, however, sound produced by females building redds has been linked to priming males for gamete release, and could be important for attracting males and synchronized gamete release (Moore and Waring, 1999). Additionally, the acoustic environment of rivers could be important for homing and navigation (Stober, 1969), but this has received little attention. One recent study found that brook trout selected locations within a river based on acoustic properties (sound pressure level) and that the different microhabitats within a river have different acoustic signatures which could be serving as proxies for habitat quality (Kacem et al., 2020). Future research should examine the importance of the river soundscape to salmonid movements and the role of sound production in communication and their behavioural ecology.

Sounds were likely produced by all three Pacific salmon species evaluated in this study; however, this study has some limitations. All sounds observed were only documented through audio files and were not confirmed with video, thus only sounds that we were confident of were used in the analysis. Sounds were selected based on repeated examination of audio files and identifying sounds first observed in series events or sounds that were not observed in all datasets. For example, the vFRT sound was observed at a higher rate during the pink and Chinook mixed school, indicating it was coming from one of the fish species (likely pink salmon) present at that time. Our method of identifying sounds is confounded by potential human error in sound identification, thus our results are preliminary but a starting point for future work on the topic. Additionally, other variables including differences in sounds produced by males and females, density dependent sounds, seasonal or other temporal variables (*e.g.*, lunar cycle), and underlining mechanisms of sounds produced could not be identified in our opportunistic study design. Further research using controlled conditions should examine mechanisms of sound production as well as other variables that might influence sound production rates to better understand sound production in salmonids.

Pacific salmon are culturally and ecologically important species in the Northeast Pacific Ocean, but many populations are declining (Riddell et al., 2018). Passive acoustic monitoring (PAM) could represent an additional monitoring technique for managers to use to track salmon populations. Underwater hydrophones, which are inexpensive and can be deployed to record in different conditions over extended time periods, could be placed on spawning grounds and used to monitor species returning to spawn (Chapuis et al., 2021; Lamont et al., 2022; Rountree et al., 2006). However, not enough is known about sound production in Pacific salmon to enable effective PAM monitoring. Understanding the different types of sounds produced by salmonids and the mechanisms that produce them could aid in providing new methods for conservation and monitoring of salmonid species around the world. Future research should examine sound produced by salmon using video confirmation to include more sounds and better distinguish different types of sounds produced (*e.g.*, air movement). Pairing underwater hydrophones with video recordings has been used to localize and confirm sounds produced by different marine fishes (Mouy et al., 2018) which could now be incorporated in research programs into fish sound production. Additionally, future work should further examine how these different sounds are produced. To date, limited research has been conducted on the mechanisms of sound production in salmonids (Neproshin and Kulikova, 1975), even though sound production mechanisms in other species of fish have been extensively studied (Demski et al., 1973; Demski and Gerald, 1972; Skoglund, 1961). Examining how salmonid species produce sounds represents a key area of research needed to fully understand sound production in salmon.

Table 4. 1. Summary of sampling effort in 2017 at Big Qualicum Hatchery. n_1 = number of days with 24-hour subsamples, n_2 = total number of 1-minute subsamples.

		Data Collected			Analysis		
Species Present	Date Range	Raceway 1 (Hours)	Raceway 2 (Hours)	Total Hours	Continuous (minutes)	1-minute subset (minutes)	Sample size (Days: n_1; Total mins processed: n_2)
Pink and Chinook	Sep 18-Oct 02	348.2	348.8	696.9	0.0	336	$n_1 = 7$; $n_2 = 336$
Chinook	Oct 03-Oct 09	177.4	167.0	344.4	1,440	162	$n_1 = 4$; $n_2 = 1,602$
Chinook	Oct 10-Oct 17	0.0	190.8	190.8	0.0	96	$n_1 = 4$; $n_2 = 96$
Coho	Oct 10-Oct 14	109.7	0.0	109.7	1,440	24	$n_1 = 2$; $n_2 = 1,464$

Table 4. 2. Surface sound series statistics by species, total series duration (start of first sound to end of last sound), duration of surface event, and latency (time from start of surface event to start of first salmon sound), duration of fish sounds, number of fish sounds and number of unique sound types.

Species	**Statistic**	**Total series duration (s)**	**Surface event duration (s)**	**Latency (s)**	**Fish sound duration (s)**	**Number of fish sounds**	**Number of unique sound types**
Chinook (n = 44)	Min	0.51	0.28	0.41	0.03	1	1
	Max	5.24	3.4	3.87	3.62	4	2
	Mean	2.23	0.92	1.43	0.80	1.36	1.18
	SE	0.18	0.09	0.13	0.14	0.1	0.06
Coho (n = 18)	Min	0.52	0.17	0.3	0.03	1	1
	Max	3.16	1.29	1.92	2.66	4	4
	Mean	1.77	0.55	1.0	0.77	1.44	1.28
	SE	0.16	0.07	0.1	0.18	0.22	0.18
Pink and Chinook (n = 22)	Min	0.92	0.1	0.19	0.03	1	1
	Max	3.01	2.02	2.38	2.07	4	2
	Mean	1.76	0.79	1.18	0.57	1.64	1.23
	SE	0.14	0.1	0.13	0.14	0.20	0.09

Table 4. 3. Sound statistics (Mean ± SE) of sounds produced by each species that occurred more than one time, ordered by mechanism then percent occurrence.

			Frequency Statistics (Hz)									
Type	**Relative Occurrence (%)**	**Sample size**	**Low**	**1st quartile**	**Peak**	**Median**	**3rd quartile**	**High**	**90% bandwidth**	**bandwidth**	**90% duration (s)**	**Duration (s)**
Chinook												
Miscellaneous air movement (AM)	30.7	96	332 ± 40	682 ± 78	829 ± 79	925 ± 129	1222 ± 186	2612 ± 334	1302 ± 210	2280 ± 307	0.25 ± 0.03	0.3 ± 0.03
Gill-bubble FRT	16.6	52	162 ± 10	453 ± 20	630 ± 45	612 ± 33	876 ± 53	2738 ± 207	1380 ± 106	2579 ± 208	0.67 ± 0.05	0.78 ± 0.06
vFRT	5.4	17	1049 ± 145	1798 ± 267	2029 ± 361	2462 ± 442	3369 ± 630	8243 ± 1814	3711 ± 792	7195 ± 1790	0.43 ± 0.23	0.51 ± 0.26
Moan	4.5	14	232 ± 34	372 ± 40	405 ± 42	419 ± 35	459 ± 32	773 ± 70	251 ± 36	541 ± 66	0.31 ± 0.07	0.39 ± 0.09
Gurgle	2.6	8	306 ± 123	527 ± 127	557 ± 139	604 ± 127	686 ± 127	1872 ± 370	633 ± 127	1566 ± 317	0.39 ± 0.11	0.48 ± 0.11
Buzz	1.6	5	1375 ± 234	3038 ± 973	3141 ± 964	3591 ± 1151	4341 ± 1429	8058 ± 4040	4144 ± 2307	6682 ± 4081	1.31 ± 0.46	1.47 ± 0.52
Growl	0.6	2	179 ± 71	281 ± 94	328 ± 141	352 ± 117	492 ± 70	1256 ± 28	680 ± 70	1078 ± 99	0.8 ± 0.42	0.89 ± 0.43
Unknown Mechanism (Pulse)	39.6	124	534 ± 34	1153 ± 89	1435 ± 117	1503 ± 119	2003 ± 156	5792 ± 5675	2360 ± 244	5258 ± 562	0.03 ± 0.002	0.06 ± 0.002
Coho												
AM	10.2	15	366 ± 64	750 ± 76	894 ± 112	891 ± 84	1072 ± 96	2076 ± 166	922 ± 75	1711 ± 185	0.24 ± 0.06	0.29 ± 0.07
Gurgle	25.9	38	266 ± 18	506 ± 23	622 ± 36	616 ± 26	759 ± 33	1343 ± 67	713 ± 45	1077 ± 61	0.34 ± 0.07	0.4 ± 0.08
Gill-bubble FRT	10.2	15	243 ± 39	544 ± 41	678 ± 70	675 ± 55	903 ± 67	2072 ± 272	1125 ± 137	1829 ± 274	0.9 ± 0.11	1.03 ± 0.13
Moan	2.0	3	361 ± 43	484 ± 41	562 ± 54	531 ± 31	594 ± 41	972 ± 174	359 ± 62	611 ± 139	0.14 ± 0.02	0.16 ± 0.02
Pulse	50.3	74	546 ± 51	1018 ± 77	1204 ± 78	1252 ± 93	1622 ± 141	4050 ± 504	1642 ± 213	3503 ± 481	0.05 ± 0.01	0.07 ± 0.01
Pink and Chinook												
AM	45.2	76	350 ± 27	883 ± 40	1135 ± 64	1136 ± 51	1594 ± 100	3841 ± 235	1943 ± 152	3491 ± 2233	0.11 ± 0.02	0.15 ± 0.02
vFRT	21.4	36	2728 ± 38	3322 ± 19	3579 ± 50	3655 ± 32	4147 ± 53	5112 ± 60	1910 ± 61	2384 ± 65	0.1 ± 0.01	0.12 ± 0.01
Gill-bubble FRT	4.2	7	251 ± 46	683 ± 108	797 ± 104	864 ± 103	1333 ± 166	3099 ± 295	1688 ± 268	2847 ± 309	0.37 ± 0.08	0.43 ± 0.09
Buzz	1.8	3	572 ± 285	1094 ± 257	1188 ± 271	1359 ± 285	1625 ± 220	3947 ± 370	1438 ± 236	3376 ± 166	0.2 ± 0.15	0.22 ± 0.16
Gurgle	1.2	2	3038 ± 161	3609 ± 281	4266 ± 187	3938 ± 281	4383 ± 117	5127 ± 76	1641 ± 234	2089 ± 237	0.25 ± 0.06	0.27 ± 0.06
FRT	1.2	2	355 ± 139	891 ± 141	797 ± 234	1266 ± 47	1781 ± 234	3161 ± 45	2297 ± 328	2806 ± 184	0.97 ± 0.75	1.09 ± 0.83
Raspy	1.2	2	1364 ± 93	2461 ± 23	3445 ± 539	3164 ± 258	3914 ± 164	8411 ± 2863	4383 ± 1148	7046 ± 2770	0.15 ± 0.03	0.18 ± 0.02
Pulse	19.6	33	687 ± 64	2040 ± 258	2278 ± 285	2588 ± 332	3486 ± 482	11298 ± 1495	4602 ± 692	10612 ± 1467	0.02 ± 0.002	0.05 ± 0.005

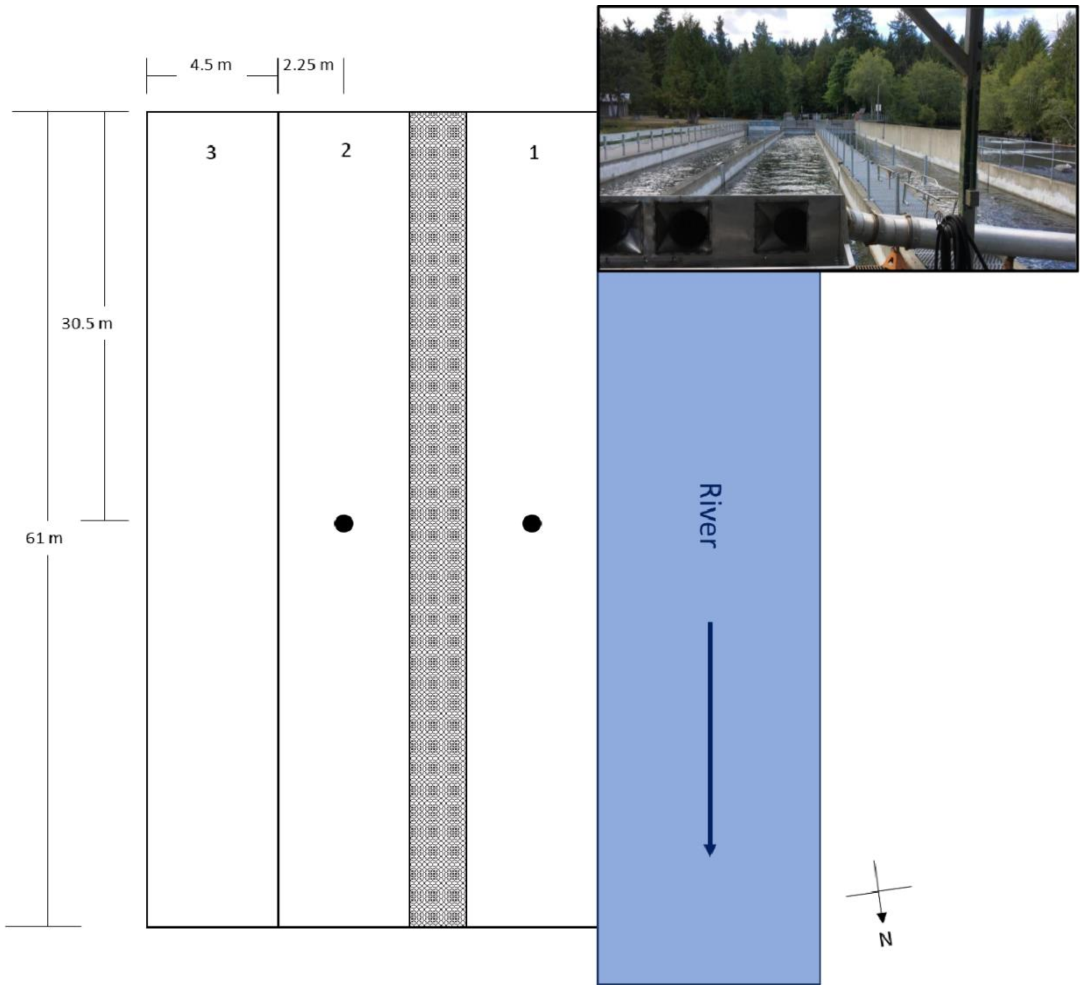

Figure 4. 1. Schematic of hatchery set up; black dot in raceway 1 and 2 represents the location of a single SoundTrap deployed in each raceway.

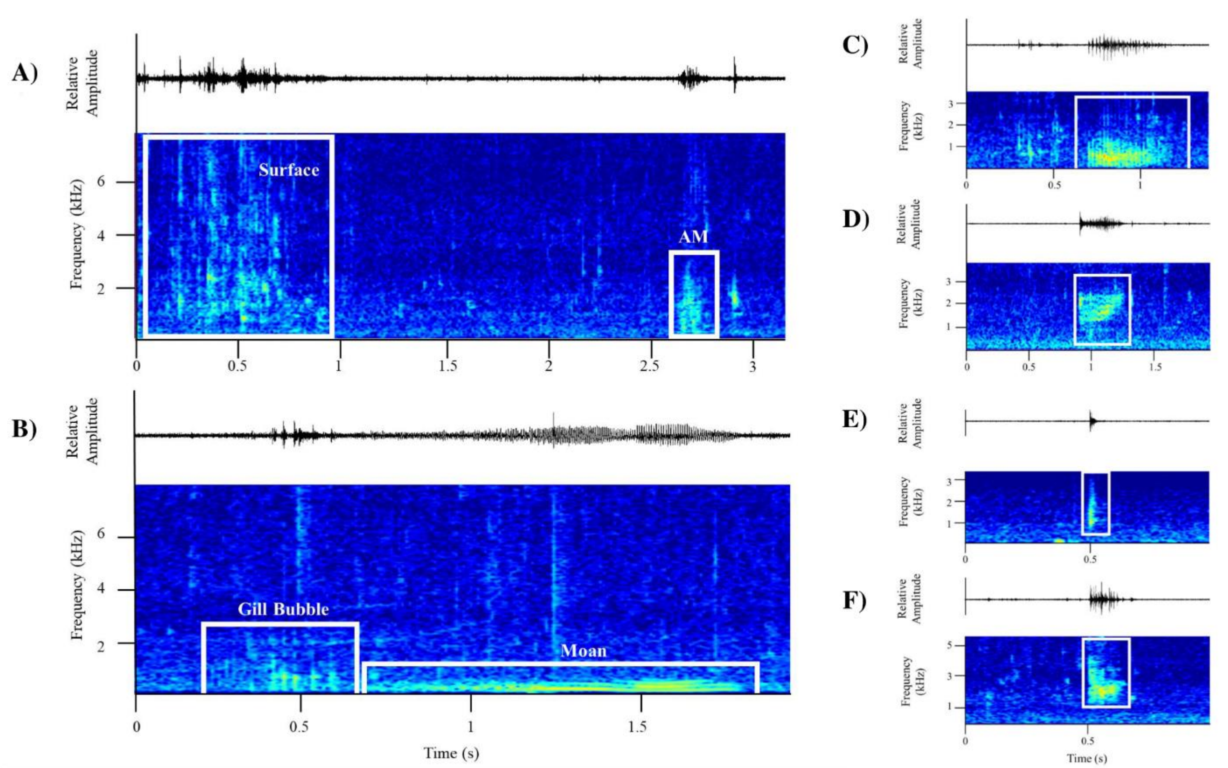

Figure 4. 2. Examples of series events and individual sounds for Chinook salmon. Top panel: the waveform filtered to each sound by bandwidth and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). To accommodate varying frequency ranges of sounds, subplots have different frequency ranges. A) Series with surface event followed by miscellaneous air movement sound. (Mm. 1) B) Short series with a gill-bubble FRT sound that connects into a moan. (Mm. 4) C) Gill-bubble FRT. (Mm. 5) D) Miscellaneous air movement. (Mm. 6) E) Pulse. (Mm. 16) F) vFRT. (Mm. 7).

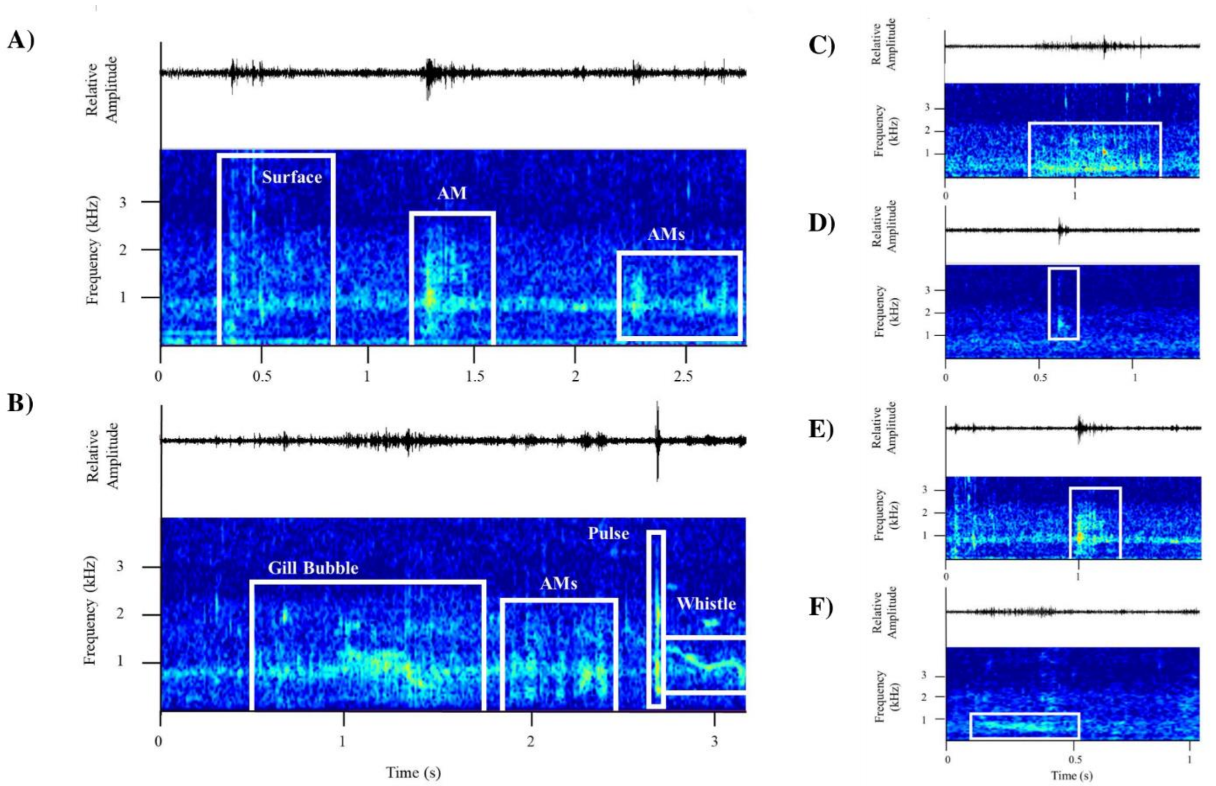

Figure 4. 3. Examples of series events and individual sounds for Coho salmon. Top panel: the waveform filtered to each sound by bandwidth and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). A) Series with surface event followed by multiple miscellaneous air movement sounds. (Mm. 2) B) A gill-bubble FRT sound that connects into multiple miscellaneous air movement sounds (AMs) and ends with a pulse sound and a whistle. (Mm. 8) C) Gill-bubble FRT. (Mm. 9) D) Pulse. (Mm. 17) E) Miscellaneous air movement. (Mm. 10) F) Gurgle. (Mm. 11).

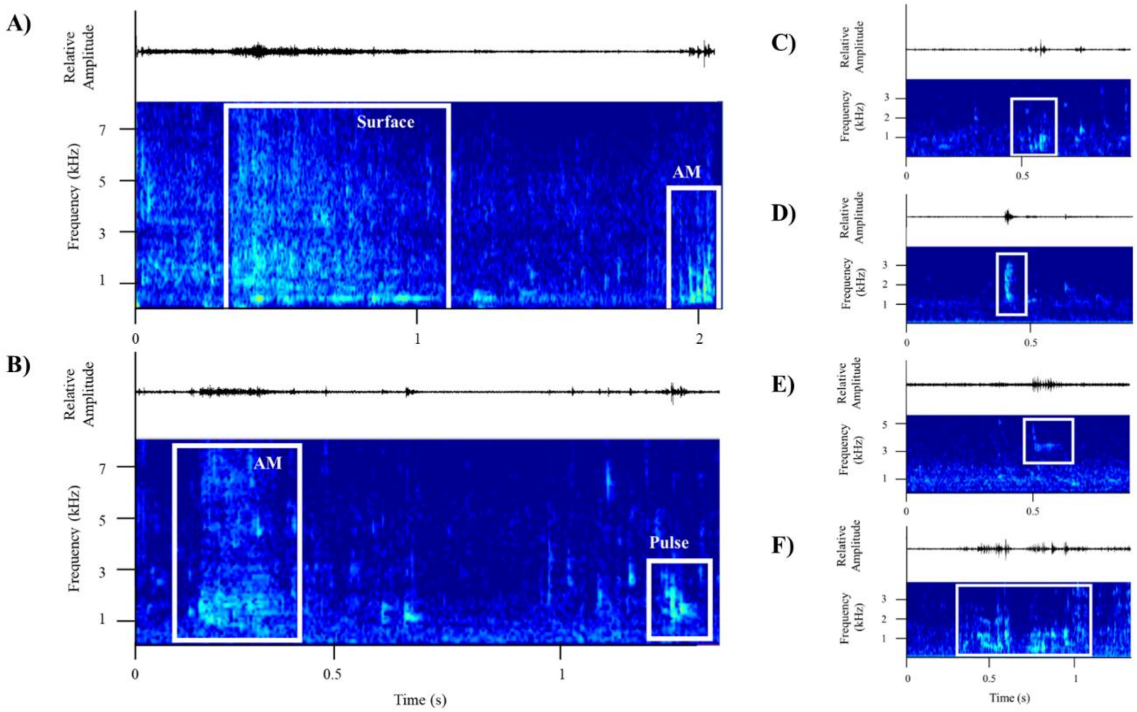

Figure 4. 4. Examples of series events and individual sounds for mixed school of Pink and Chinook salmon. Top panel: the waveform filtered to each sound by bandwidth and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). To accommodate varying frequency ranges of sounds, subplots have different frequency ranges. A) Series with surface event followed by a miscellaneous air movement sound. (Mm. 3) B) Short series with a miscellaneous air movement followed by pulse sound. (Mm. 12) C) Miscellaneous air movement (Mm. 14) D) Pulse. (Mm. 18) E) vFRT (Mm. 15) F) Gill-bubble FRT (Mm. 13).

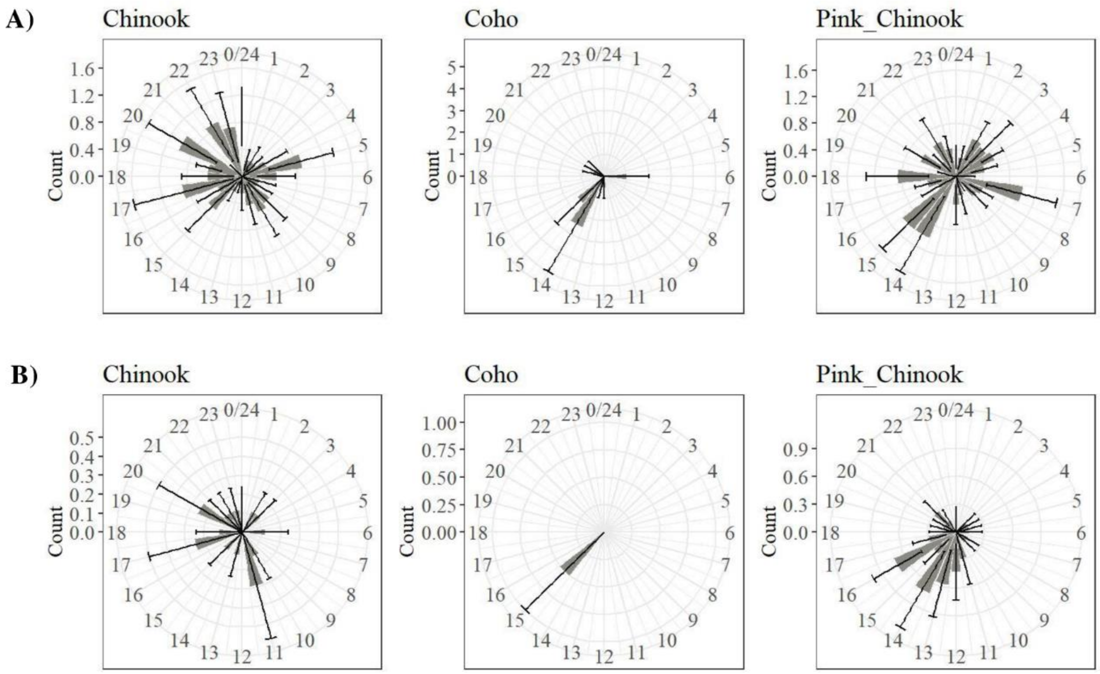

Figure 4. 5. Circular bar graphs of mean and standard error of counts of sound production by hour of the day in each grouping of salmon, Chinook only ($n_1 = 8$), Coho salmon ($n_1 = 2$), Pink and Chinook mixed ($n_1 = 7$). A) Miscellaneous air movement sounds. B) vFRT sounds.

Chapter 5- Behavioural responses to shipping noise and terminal migration movements of Chinook salmon in Cowichan Bay, British Columbia

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5.1 Abstract

The increase in human-generated sound over the last 60 years has led to concerns regarding the impacts of shipping noise on marine species. Two important marine species in the Northeast Pacific are declining, southern resident killer whales (*Orcinus orca*) and their main prey, Chinook salmon (*Oncorhynchus tshawytscha*). We know that killer whales change their behaviour in the presence of ships, but no work has been done on Chinook salmon. An acoustic telemetry study, together with soundscape monitoring, was conducted to examine potential changes in behaviour of acoustically tagged Chinook as they encounter shipping noise. Seventeen acoustic telemetry receivers and eight underwater hydrophones were deployed in Cowichan Bay, British Columbia, Canada over three years (2019, 2020, and 2022). Depth and activity levels of adult Chinook salmon were monitored using acoustic tags (n = 55), and were modelled against environmental conditions (*e.g.*, tides) to understand general movement and behaviour. Underwater sound pressure levels, based on concurrent soundscape recordings, were then added to the model to assess potential responses to ship noise. Chinook salmon had strong general diel detection and behaviour patterns, with increased detections in Cowichan Bay and salmon were shallower during the day, while a decrease in detections and salmon being deeper overnight. Additionally, when sound levels were included in the model, Chinook salmon were found to be deeper at higher sound pressure levels (> 120 dB re 1 µPa; 20-24000 Hz) and exhibited significant changes in depth (shallower) and activity levels (higher activity levels) when one AIS-equipped vessel was present, demonstrating the first data on Chinook salmon responses to anthropogenic noise. These data provide novel insights into the behaviour of adult Chinook salmon and crucial information on the impacts of anthropogenic noise on this key species.

5.2 Introduction

Catch rates of Chinook salmon (*Oncorhynchus tshawytscha*) in the Northeast Pacific Ocean dramatically declined starting in 1975 (Riddell et al., 2013). Chinook salmon stocks around the Salish Sea (waters located between southern British Columbia, Canada and northern Washington State, United States which includes the Strait of Georgia, the Strait of Juan de Fuca, and Puget Sound) saw declines in smolt-to-adult return rates for ocean-type Chinook salmon between 1970 and 2000 but have remained consistently low (~1%) since 2000 (Welch et al., 2021). Additionally, marine survival for Chinook salmon was the lowest in the 1990s and 2000s compared to higher marine survival rates found in the 1970s and 1980s (Riddell et al., 2013). Not only are marine survival rates low, but Chinook salmon have also been decreasing in size-at-age since the late 1970s (Ohlberger et al., 2019), leading to decreased fecundity (Malick et al., 2023). These ongoing declines result in fewer and smaller individuals returning to spawn making it more challenging for the stocks to recover.

Anthropogenic factors have been negatively affecting Chinook salmon populations for decades (Cao et al., 2022; Nehlsen et al., 1991). Typically, these factors are related to pollution (Cao et al., 2022), habitat loss (Nehlsen et al., 1991), or hatchery enhancement (Levin et al., 2001; Mcmillan et al., 2023) but, another potential influence is from underwater noise, which has received little attention (van der Knaap et al., 2022). Underwater noise from vessels has been shown to impact other fish species by altering movement (Ivanova et al., 2020), foraging (Voellmy et al., 2014a) and antipredator behaviours (Voellmy et al., 2014b), which can have population-level consequences (Soudijn et al., 2020). Research into the impact of boat noise on salmon has been gaining interest, with one study evaluating changes in behaviour on juvenile pink (*Oncorhynchus gorbuscha*) and chum salmon (*Oncorhynchus keta*) when boats pass by (van der Knaap et al., 2022). Van der Knaap et al (2022) found 50% of salmon behaviourally (school cohesion, school orientation, and swimming speed) responded to boat noise above 140 dB re 1μPa (10-20,000 Hz).

While movement patterns of juvenile Chinook salmon have been explored around the Salish Sea (Arostegui et al., 2017a, 2017b; Chamberlin et al., 2011a, 2011b; Kagley et al., 2017; Smith et al., 2015), the movement patterns of adult Chinook salmon have received limited attention in the literature (Candy and Quinn, 1999; Courtney et al., 2021). Courtney et al. (2021) tracked adult Chinook salmon off the Gulf of Alaska using PSAT tags and examined movement patterns and depth distribution. Most salmon remained around Cook Inlet, where tagging occurred but a few travelled south down the coast of Alaska, travelling between 7 to 56 km each day (Courtney et al., 2021). Additionally, depth data showed that Cook Inlet residents spent the majority of their time (48%) near the surface (< 10 m) but did dive to depths between 30 and 50 m often, while transient salmon that left the bay tended to dive deeper (> 200 m) and spent more time at depths greater than 100 m (Courtney et al., 2021). Another study on Chinook salmon movements in Cook Inlet using acoustic tags found similar results, with Chinook salmon being near the surface (4.85 m, median depth) and travelling close to shore (~2.5-7.5 km offshore; Welch et al., 2013). Welch et al. (2013) also showed differences in travel time between movement patterns in the ocean and river, with salmon moving slower in the ocean, indicating potential staging behaviours. Movement and depth patterns have also been explored in Chinook migrating along the coast of British Columbia (Candy and Quinn, 1999). During their spawning migration, Chinook salmon tracked in Johnstone Strait were found to travel at speeds between 0.7 to 2.7 km/hr and at a mean depth of 70 m (Candy and Quinn, 1999). Candy and Quinn (1999) also observed that migrating salmon were found close to shore at a distance to shore of between 0.2 and 1.7 km.

Chinook salmon are a crucial component of the Northeastern Pacific Ocean ecosystem, but alterations of their behaviour in the presence of vessel traffic has yet to be evaluated. Impacts of vessels have been shown to alter behaviour in both resident killer whales (*Orcinus orca*; *e.g*., Lacy et al., 2017; Williams et al., 2002), one of their major predators (Ford and Ellis, 2006), and Pacific herring (*Clupea pallasii*; van der Knaap et al., 2022), one of their principal prey items (Osgood et al., 2016). So, understanding the behavioural changes exhibited by Chinook salmon in response to shipping noise would increase our knowledge of ecosystem-wide impacts of elevated noise levels and potentially provide information on how predatory-prey relationships might be affected. Furthermore, little research to date has evaluated terminal migration movements and behaviours of Chinook salmon (Candy and Quinn, 1999; Welch et al., 2013; Courtney et al., 2021), with a specific focus on understanding how environmental or predation pressures could be altering their movements and behaviours. This study aimed to understand the general movement and behaviour patterns of adult Chinook salmon before in-river migration and document potential changes in their behaviour from vessel noise.

5.3 Methods

5.3.1 Study location

Cowichan Bay, British Columbia, is an industrialized bay located on the east coast of Vancouver Island (48° 44.975' N, 123° 35.922' W). The bay is approximately 4 km long and 2 km wide and is at the mouth of the Cowichan River. The Cowichan River originates at Cowichan Lake and is bifurcated into two arms (north and south) that connect the estuary to the main channel (Figure 5.1). Additionally, Cowichan Bay has an elevated amount of anthropogenic noise from recreational boats (two marinas), and commercial vessels, including large vessels anchoring at one of six anchoring locations as well as tugboats bringing logs into the estuary to be stored prior to processing at the mill located in the Cowichan estuary.

The Cowichan River has a fall run population of Chinook salmon that moves upriver in early October. However, local knowledge suggests that Chinook salmon remain in the bay for a few weeks (mid-August - early October) before moving up the Cowichan River to spawn. The Cowichan River Chinook salmon stock is the indicator stock for the lower Strait of Georgia (Tompkins et al., 2005) and as such is monitored with a Passive Integrative Transponder (PIT) tag research program managed by Fisheries & Oceans Canada (DFO), British Columbia Conservation Foundation and Cowichan Tribes (Cantera and Damborg, 2023). Cowichan Tribes also manages a community hatchery located approximately 2 km upriver, and almost all Chinook salmon (~100% mark rate) released are marked with a coded-wire tag and have their adipose fin removed (Tompkins et al., 2005).

5.3.2 Experimental design

Twelve VR2W and three VR2AR omnidirectional acoustic receivers (VEMCO, Bedford, NS, Canada) were deployed in a variety of environments throughout Cowichan Bay, estuary, and river between August and November 2019 (Figure 5.1, Table 5.1). Nine receivers (6 VR2W and 3 VR2AR) were deployed in Cowichan Bay, all but one receiver was attached to polysteel rope connected to a “deck block” with 200 mm trawl floats to keep the receiver vertical. Five of those receivers were deployed in shallow water (18 m) and anchored to shore with leaded line, while three were deployed deep (> 30 m) with acoustic releases. The final receiver was secured to a leaded line that was connected to a 6.8 kg cannonball and attached to a mooring buoy and deployed at 21 m. In 2020, an additional two VR2AR omnidirectional acoustic receivers were deployed outside Cowichan Bay with acoustic releases. In the final year (2022) all acoustic receivers (VR2W and VR2AR) were deployed in the same locations. The remaining six VR2W acoustic receivers were deployed in the estuary (2) and river (4) in all three years. Each receiver was attached to rebar with cable ties and mounted inside a cinder block.

SoundTrap underwater hydrophones (STD300 and STD300HF; Ocean Instruments, New Zealand, system gain: -171.7 to -179.4 dB re FS/µPa, flat frequency response from 10 Hz to 72 kHz, self-noise < 34 dB re 1 µPa above 2 kHz) were used to collect sound pressure level data in the bay. They were deployed at between three and eight locations within Cowichan Bay depending on the year (Table 5.1) and were attached 30 cm above the VEMCO acoustic receiver at those locations. All hydrophone systems were set to sample at 48 kHz at 16 bits. Two recorders, Front Mid Bay and N Bay Opening, followed a duty cycling approach, recording for 15 minutes every hour, while the rest maintained continuous recording. In 2022, only four SoundTrap hydrophones were deployed, and all were duty cycled for 15 minutes every hour. Prior to deployment, the SoundTraps underwent manufacturer calibration at 250 Hz using a B & K 2236 pistonphone set at a source level of 120 dB re 1 µPa.

5.3.3 Tagging

Adult Chinook salmon were captured individually with standard salmon angling gear in Cowichan Bay, British Columbia. The salmon were quickly transferred to a large cooler of seawater maintained within 2 degrees Celsius of ambient seawater. Fish were held in the water in an inverted position in a dark-coloured Colman cooler (Xtreme Marine Pro, 100 Quart) to reduce stress (Gaffney et al., 2016). Prior to proceeding with tagging, each fish was assessed for viability for tagging based on reflex tests. The reflex tests included eye tracking, tail reflex, ventilation, and ability to right self (orientation; Cook et al., 2018). If the fish failed any of these tests it was deemed untaggable, and euthanized (University of Victoria Animal Use Protocol: 2019-013). The sampling protocol proceeded when the fish had functional reflexes.

For each fish, fork length and circumference were measured, then a small fin clip was taken for genetic stock identification. A fin clip was taken from the adipose or caudal fin, depending on origin, wild (adipose fin present) or hatchery (no adipose fin). Then, five scales were removed from just above the lateral line for aging. While samples were being collected, a variety of condition factors were assessed, including fin damage, wounds, scale loss, sea lice presence, origin (adipose fin present/not), degree of bleeding (0-3; no bleeding, light, moderate, significant), maturity (0-3; no colouring, light, moderate, significant), and sex. A Passive Integrative Transponder (PIT) tag was injected intraperitoneally in front of the pelvic girdle using a new, sterile 12-gauge needle (Biomark; Boise, ID: FDX-B 12mm) for use in a larger study assessing terminal survival (Atkinson et al., 2023) but those data are not used in this chapter. An acoustic tag (VEMCO; Bedford, NS; V13AP-1x, 69 kHz) was inserted into the salmon's stomach through the fish's mouth using a plastic plunger. Both the tag and plunger were lubricated with glycerin to ensure no damage was done to the esophagus following procedures in Burger et al. (1985). The acoustic tag was set to randomly ping between 30-50 seconds and was equipped with two behaviour sensors (depth and acceleration), which were alternated at a rate of 1:1. Depth values (range: 0-136 m, accuracy ± 6.8 m, resolution = 0.6 m) were generated using a pressure sensor at one end of the acoustic tag, and acceleration values (range: 0-4.9 m/s^2) were calculated by the root mean squared (rms) value of acceleration on three axes (x, y, z), sampled at 10 measurements per second for 20 seconds collected immediately after the depth reading is transmitted. The acceleration values were used to assess general activity of the fish, however, previous research has shown that accelerometer readings have a significant positive correlation with tail beat frequency in sockeye salmon (*Oncorhynchus nerka*; Wilson et al., 2013). Additionally, higher readings (>3 m/s^2) have been shown to represent burst activities in fish

(Murchie et al., 2011), representing the potential to document fast movements. Since in this study we did not evaluate the connection between swimming speed and the accelerometer reading, accelerometer readings will be referred to as activity levels for the remainder of the chapter.

A pilot study to assess the rate of retention of the acoustic tag on terminal salmon that were gastically tagged was conducted in 2018, which found a 100% retention (Appendix C). Additionally, due to the low number of tags available for the study, not all salmon captured were acoustically tagged. Only salmon that were larger than 50 cm fork length and had no to little bleeding (0 or 1) were acoustically tagged. Salmon that did not meet these requirements were only PIT-tagged for the larger terminal survival study (see Atkinson et al., 2023).

5.3.4 Analysis

Data from each receiver were downloaded using VEMCO VUE software and compiled into a single data set for each year which were then imported into RStudio (RStudio, 2020) and combined for analysis. Mean and standard error (SE) for fish metrics (fork length and circumference) and ratios for fish sex, origin and stock were tabulated for each year of tagging. Genetic stock identification was conducted on each fin clip to obtain the probability of which of the North American Chinook salmon stocks the fish belonged to; methods for this procedure are outlined in Beacham et al. (2021). Samples from all three years were run as a single group using the cBayes program (Neaves et al., 2005), and probability estimates were found following methods outlined in Pella and Masuda (2001). Only salmon with a probability over 75% were considered to be from the Cowichan stock. Data were not normally distributed, so non-parametric Kruskal–Wallis ANOVAs with Dunn’s post hoc tests were used for analysis of differences among years for fish metrics.

Detections for the first 15-minutes after tagging were extracted for each individual fish and examined for post-tagging behaviours (*e.g.*, increased activity, deep dives). Marine mortality was assessed by examining the depth and activity values to see if the tag was on bottom and not moving (activity values < 0.05 m/s^2), or if a tag was not detected leaving Cowichan Bay (last detection on one of the acoustic receivers at the opening of Cowichan Bay) and disappeared between two receivers within the array, it was considered to be 'deceased'. Fish that were found to be not moving and on bottom and disappeared within 24-hours after tagging were considered 'tagging-related mortalities' (Candy et al., 1996). Each fish that was considered 'alive' after 24-hours but was later found on bottom was examined to determine if prior behaviours could indicate a predation event (*e.g.,* changes in activity levels and depth). Fish that were found to be moving upriver (detected on estuary receivers) but then disappeared between two receivers were considered 'deceased' and counted as freshwater mortality.

Since the environmental conditions in Cowichan Bay could potentially influence the behaviour of tagged salmon, various environmental variables for Cowichan Bay were collected and added to the dataset. Time of day (hour) and day of the year (Julian) were extracted for each detection. Tide height above sea level (m) data were obtained from the Cowichan Bay station (07310) on the Government of Canada website in 15-minute blocks and tide direction (ebb or flood) was added based on whether the previous height was higher or lower than the next time point. River temperature (degree Celsius) and river discharge were obtained from the Water Survey of Canada Station (08HA011) in Duncan, British Columbia, in one-hour blocks (temperature) and 5-minute blocks (discharge). Each detection from one of the bay receivers was then matched to the closest block for tide data, while detections from the estuary or river receivers were matched to the temperature and discharge data.

Additionally, sound and vessel variables were added to the main dataset from the bay telemetry receivers to be used in the model to understand the changes in behaviour with vessels. Data on vessel numbers, location and movements were obtained from the Canadian coastguard. Vessels embarking on international voyages with a gross tonnage over 300, those undertaking domestic voyages with a gross tonnage exceeding 500, and all passenger vessels are mandated by the International Maritime Organization (IMO; 2015) to report their position and identification data using the Automatic Identification System (AIS). AIS position data, with an accuracy of ± 1 m, were recorded at 5-minute intervals for all AIS-equipped vessels within a radius of approximately 5 kilometers (km) around Cowichan Bay during the months of August through October in 2019, 2020, and 2022. The number of AIS vessels were enumerated for a 1 km radius around each receiver (estimated range of acoustic tags) and matched to individual detections in the main dataset. Most vessels included in the data were required to track their position (IMO, 2015), but a small percentage (8.5%) were recreational vessels or an unknown vessel type. Noise levels were obtained from the SoundTrap recordings analyzed in MATLAB R2018b, to aid in understanding the impact from untracked vessels (recreational vessels). Sound Pressure Levels (SPL) were calculated over the full broadband range (20-24,000 Hz) for the minute prior to each individual detection to match the collection window for the activity and depth sensors.

Detection patterns within Cowichan Bay and River were first examined descriptively and then statistically modelled. Only salmon found to be of Cowichan origin through genetic stock identification (GSI), hereafter Cowichan Chinook, were used to understand the movements and behaviours of tagged salmon in Cowichan Bay (n = 35). Patterns of Cowichan Chinook movements into and out of the estuary and lower river were tabulated by year to understand annual variation in river migration patterns. Detection and behavioural (activity level or depth) patterns within Cowichan Bay and River were modelled using hierarchical generalized additive models (GAMs; Pedersen et al., 2019).

Detection patterns were examined by counting the number of individual salmon at each station for each hour of the day. To account for variation in the time spent in the bay and river, each fish was only available to be counted from its tagging date to river entry (bay) and river entry to last detection in the river (river). Detections of fish were modelled with smoothing terms for hour of the day, which changed by station, and day of year, which changed by year. Hour of the day was modelled with a cyclic cubic spline to ensure that the start and end matched (0 hour and 23 should be similar values), because hour of the day is cyclical. Year and Station were also included as parametric terms. Since fish could only be detected at one station at a time, the data have a high proportion of zeros; therefore, a zero-inflated Poisson family was used (ziP: Wood, 2006). Models (gam) were run using the mgcv package (Wood, 2006) and used a restricted maximum likelihood (REML).

Behaviour changes were also modelled with environmental variables (time of day, day of year, tide height, and tide direction) and then modelled with opportunistic encounters with vessels (AIS vessels, Sound Pressure Levels). Both models (depth and activity levels, modelled separately) were based on *a priori* hypotheses that behaviour would change based on the environmental and vessel variables, with no model selection conducted. For example, other species of fish have a daily pattern of being deeper during the day and shallower overnight (Gauthier and Rose, 2002), and many fish will respond to noise similar to predators (Jacobsen et al., 2014), so salmon would be expected to dive deeper (Yano et al., 1984). Smoothing terms were included in the model for time of day, day of the year, tide height by direction, and sound pressure level, with additional parametric terms for tide direction, and number of AIS vessels. Fish ID and Station were used as random variables in the model to account for individual variation among fish and at different locations around Cowichan Bay. Similar to the movement models, hour of the day was modelled with a cyclic cubic spline. Smoother knots (‘k’) were set for each variable individually based on the potential degree of responsiveness to the variable (Lennox, R., personal communication). All environmental variables (tide height, hour of day) were set to k = 5, but since there could be more variation in response to sound pressure level (k = 6) and day of year (k = 20), the number of knots were increased. Similarly, behavioural changes (activity level or depth) within Cowichan River were also modelled using hierarchical generalized additive models (GAMs) with environmental variables (time of day, day of year, river temperature, river discharge), following the same design as behaviours in the bay. Since depth and activity values were non-normally distributed with only positive values, depth and activity levels were modelled with a gamma distribution with a log link. Models (bam) were run using the mgcv package (Wood, 2006) using a restricted maximum likelihood (fREML) to account for the large number of detections used in the models and discrete = TRUE to apply an autocorrelation structure. Models were checked using the gam.check function in the mgcv package (Wood, 2006) to confirm k-values were adequate. All statistical tests were performed in RStudio version 4.2.3 (R, 2021).

5.4 Results

5.4.1 Fish metrics

Each year a similar number of salmon were acoustically tagged (2019: n = 17; 2020: n = 19; 2022: n = 19; Table D5.1). Tagged salmon had a mean (± SE) fork length (FL) of 765.7 ± 12.1 mm and mean (± SE) circumference of 450.3 ± 6.5 mm in 2019. Fork length and circumference decreased to 716.8 ± 20.6 mm and 417.2 ± 12.0 mm in 2020, then decreased again in 2022 (FL = 703.7 ± 14.8 mm; circumference = 391.6 ± 8.7 mm). Tagged salmon in 2022 had significantly shorter fork lengths compared to 2019 salmon (Kruskal–Wallis, df = 2, $p = 0.009$), but not to 2020 (Kruskal–Wallis, df = 2, $p = 0.24$), no difference was observed between 2019 and 2020 salmon (Kruskal–Wallis, df = 2, $p = 0.22$). All years were statistically different from each other for circumference (Kruskal–Wallis, df = 2, $p < 0.05$). The sex ratio of tagged salmon was 6 females, 5 males, and 6 were unknown for 2019. For 2020 there were 10 females and 9 males tagged, while in 2022 sex markers were not analyzed resulting in all 19 tagged fish being of unknown sex. The majority of tagged salmon originated from the Cowichan River stock at 76.5% (2019), 79.0% (2020) and 73.7% (2022). Additionally, most of the tagged Cowichan Chinook were of wild origin, with only 2 from both 2019 and 2020 (none from 2022) originating from the hatchery.

Individually tagged salmon were tracked for up to 15 minutes post-tagging. In 2019, all acoustically tagged salmon (17) were tracked for up to 15 minutes post tagging, but in 2020, fifteen of nineteen were tracked and only ten of nineteen were tracked in 2022. After initial burst behaviour, all individuals displayed similar activity levels of around 1 m/s^2 (Figure E5.1). Immediately upon release, five Chinook accelerated away from the boat; two of these displayed activity levels above 4 m/s^2, while the other three withdrew at about 3 m/s^2, likely indicating burst behaviour away. Following any burst behaviour, all fish stabilized between 0.5-2.0 m/s^2. Three depth profiles were observed. Half of the tracked salmon (n=21) displayed expected post-tagging behaviour by diving deep immediately, but twenty Chinook remained near the surface, and one started to dive, then returned to the surface (Figure E5.2).

5.4.2 Mortality

Potential post-tagging mortality was also determined for all tagged individuals. In 2019, only one salmon (ID 7940) was detected on the bottom of the bay (52 m) approximately 3 hours after tagging, resulting in a tagging mortality estimate of 6%. However, in 2020 and 2022, post-tagging mortality was higher with 16% in 2020 and 10% in 2022. In 2020, three salmon (IDs 10584, 10620, and 12176) had detections only on the day of tagging (less than 24 hours after tagging) and were later found not to be moving and on the bottom (58-145 m) during a dead tag sweep on November 11, 2020. Additionally, in 2020, one tagged salmon (ID 10608) showed a change in behaviour the day after tagging. Activity levels significantly increased (Mann-Whitney-Wilcoxon t Test, $W = 706.5$, $p < 0.001$) from a mean ± SE of 0.8 ± 0.04 m/s^2 to a mean of 2.45 ± 0.07 m/s^2 from five hours after tagging until the tag was found on bottom (50 m) two days after tagging, and depths values also significantly increased (Mann-Whitney-Wilcoxon t Test, $W = 8487.5$, $p < 0.001$) from 7.4 ± 0.9 m to a mean of 12.5 ± 0.75 m. The change in behaviour likely indicates a pinniped predation event. However, since this predation event was less than 24 hours after tagging, handling, and tagging stress likely contributed to this event. Finally in 2022, two salmon (IDs 3958, 3982) were detected on bottom (3-63 m) in Cowichan Bay during the yearly dead tag sweep on December 2nd and 8th, but both salmon were only detected on the array for less than three hours before disappearing. Overall tagging mortality was 10.9% (6/55) when combining all three years of data.

Freshwater mortality was determined based on the number of individuals detected in the river compared to detections at the counting fence. Freshwater mortality was estimated to be 50% (6/12) in 2019, with one individual dying in the estuary under the docks (ID 8218) on the south side of the estuary and another (ID 7954) in the south arm of the Cowichan River. Salmon 7954 perished within range of an acoustic receiver in the lower river (south), with activity levels maxing out (4.9 m/s^2) multiple times before being found on bottom of the river (0.7 m) and is assumed to be a pinniped predation event. Four other individuals entered the estuary or river, but their fates were not confirmed. One tagged salmon (ID 4636) was detected in the south side of the estuary but never made it into the lower river, while two salmon (IDs 7958, 8212) were last detected in the south arm of the lower river. The final salmon (ID 7944) was detected in the river's main stem (Major Jimmy) but never made it to the counting fence at river km 7. In 2019, a total of six individuals (50% freshwater mortality) crossed the counting fence between September 13 and October 18, 2019. In both 2020 and 2022 all tagged salmon (2020 = 11; 2022 = 8) that entered the river survived to the counting fence (0% freshwater mortality), crossing the counting fence between September 14 and October 9, 2020, and October 2 and October 28, 2022.

5.4.3 Behaviour and Movement Patterns in Cowichan Bay

A strong increase in detections was observed daily at most stations within Cowichan Bay (Figure 5.2). Inside Cowichan Bay there was a significant increase (> 0.5 fish per hour) in detections of fish per hour during the day, peaking around 1200 (Front mid Bay: Chi.sq = 296.6, $p < 0.001$; Marina: Chi.sq = 153.1, $p < 0.001$; N 2A: Chi.sq = 221.6, $p < 0.001$), which became very low overnight (<0.2 fish per hour; between 2000 and 0500), representing a 4 to 12-fold increase in fish detected during the day depending on the station. A similar pattern was observed in the middle bay receivers (Genoa Bay: Chi.sq = 94.1, $p < 0.001$; Mid Bay: Chi.sq = 227.2, $p < 0.001$; South Mid bay: Chi.sq = 75.8, $p < 0.001$), with increased fish detected per hour during the day (1100-1600) but only some detections overnight (<0.3 fish per hour; 2100-0800). Receivers located at the opening to the bay had higher fish detected per hour overall compared to the middle and inside locations, but less of a diel pattern. Fish detected per hour significantly increased during the day (peak = 1800) at the Mid bay opening (Chi.sq = 48.2, $p < 0.001$) and South Bay opening (Chi.sq = 70.0, $p < 0.001$;peak = 1400); however, the North Bay opening (Chi.sq = 10.5, $p < 0.001$) was more consistent throughout the day but still had a significant increase in the afternoon (peak = 1800). Once outside Cowichan Bay, fish detected per hour remained lower than the bay receivers (< 0.2 fish per hour), but Sep Point 1 which is the closest to Cowichan Bay saw a significant increase in fish detected around 05:00 (Chi.sq = 17.4, $p < 0.001$), detection at Sep Point 2 remained around 0.1 fish detections per hour throughout the day (Chi.sq = 4.8, $p = 0.05$).

Multiple environmental predictor variables had a significant relationship with Chinook salmon depth. Hour of the day had a significant relationship ($F = 21610.3$, $p < 0.001$) with depth, where salmon were found significantly deeper overnight (predicted depth ~30 m) and shallower (predicted depth ~ 13 m) during the day (Figure 5.3A). Julian day was also significant with Chinook salmon depth ($F = 144.5$, $p < 0.001$), with a general trend in predicted depths increasing through time (Figure 5.3B). Environmental variables were also important, with tide height having a negative relationship with depth (Ebb: $F = 44.8$, $p < 0.001$; Flood: $F = 31.3$, $p < 0.001$); when the tide was higher salmon were shallower (Figure 5.4A), but the direction parametric coefficient was not significant ($t = 0.3$, $p = 0.75$), demonstrating a similar relationship with depth for both directions (ebb and flood). Spatial distribution also had a significant relationship with depth in Cowichan Chinook ($F = 1055.2$, $p < 0.001$), with salmon found deeper outside Cowichan Bay at stations Sep Point 1 and 2 (predicted depth ~ 30 m), and shallower at stations Front Mid Bay and N 2A located near the estuary (predicted depth < 17 m). Salmon ID was also significant ($F = 235.1$, $p < 0.001$), with individual variation observed. For example, salmon 10612 and 12178 were found to be the deepest (predicted depth > 35 m), while salmon 10624 and 10614 were the shallowest (predicted depth < 7 m), all other salmon depths were somewhere in between these four fish.

Significant relationships were found for multiple predictor variables with activity levels for Cowichan Chinook. Salmon displayed significantly ($F = 2501.4$, $p < 0.001$) faster activity levels overnight (> 1 m/s^2) and slower activity levels (< 1 m/s^2) during the day (Figure 5.3C). Julian day of the year was significant with activity levels ($F = 21.9$, $p < 0.001$), with salmon moving faster towards the end of the study (Figure 5.3D). Tide height was an important environmental variable, with a strong negative relationship with activity levels (Ebb: $F = 10.8$, $p < 0.001$; $F = 16.3$, Flood: $p < 0.001$), where when the tide was low salmon had higher activity levels, and at high tide, salmon moved less (Figure 5.4C). The direction of tide (ebb or flood) also had a significant relationship with activity level ($t = -4.0$, $p < 0.001$), with salmon being less active on the flood tide (Figure 5.4D). Spatial distribution was also found to have a significant relationship with activity levels in Cowichan Chinook ($F = 334.1$, $p < 0.001$), with salmon moving slower at the opening to Cowichan Bay (N Bay Opening, S Bay Opening, Mid Bay Opening; predicted activity level ~ 0.8 m/s^2), and fastest at stations near the estuary (Front Mid Bay, Marina, and N 2A; predicted activity level ~ 1.0 m/s^2). Salmon ID was also significant ($F = 349.4$, $p < 0.001$), with variation observed among individuals. For example, salmon 238 was found to be moving the fastest (predicted activity level = 1.2 m/s^2), while salmon 3972 was the slowest (predicted activity level = 0.4 m/s^2), all other salmon fell somewhere in between these two fish.

Sound pressure levels and number of AIS vessels present had a significant effect on the depth of Chinook salmon. The broadband sound pressure level (SPL_{rms} 20-24000 Hz) for the minute prior to the detection showed a significant relationship with the depth of Cowichan

Chinook ($F = 8.6$, $p < 0.001$). At higher sound levels (> 120 dB re 1µPa), Cowichan Chinook were deeper (> 30 m; predicted depth) compared to lower sound levels (90-120 dB re 1µPa; Figure 5.5A). Additionally, the number of AIS vessels within 1.0 km of each receiver was significant when one AIS vessel was present ($t = -2.1$, $p = 0.03$) and was approaching significance when there were two ($t = 2.0$, $p = 0.051$) (Figure 5.5B), however, there was a small number of times when two AIS equipped vessels were present at the same station (8), compared to when there were one (1060).

Broadband sound pressure levels ($F = 1.1$, $p = 0.42$) did not have a significant effect on the activity levels of Cowichan Chinook (Figure 5.5C). Salmon remained around 1 m/s^2 (predicted) at all sound pressure levels examined, but even though the difference was not significant there was a small decline in activity levels at higher sound pressure levels. However, the number of AIS vessels present did have a significant effect on activity levels; when one vessel was present ($t = 4.9$, $p < 0.001$), salmon had higher activity levels compared to when there were none, but there was no difference when two ($t = -0.1$, $p = 0.92$) were present (Figure 5.5D). However similar to depth, there were a small number of times (5) when two AIS vessels were present at the same station compared to when there was only a single vessel (1100).

5.4.4 Movement and behaviour patterns in the Cowichan Estuary and River

Chinook salmon movement into the Cowichan River also varied among the three tagging years. Only in 2019 did tagged salmon ($n = 6$) move into the estuary and then move back out of the freshwater system and back to the bay (marine environment), with four of the six fish moving into the south arm and back out while the other two made similar movements using the north arm. In all years tagged salmon ($n = 10$) moved into the lower river and then moved back to the estuary, with varying degrees. Cowichan Chinook moved the most in 2019 ($n = 6$), with one individual moving in and out of the estuary four times and three others moving between the lower river and estuary two times. In 2020, only three salmon moved out of the lower river and back to the estuary, and only one salmon exhibited this behaviour in 2022. No salmon in all three years moved from the river's mainstem back to the lower river, indicating that once a certain point in the river was crossed, any subsequent movement was only upstream.

Daily detection patterns also varied by location throughout the Cowichan estuary and river, and overall, average fish detected per hour in the river/estuary were lower than in the bay receivers (Figure 5.6). In the estuary, the south arm had a significant diel pattern with fish detected per hour increasing in the afternoon (Chi.sq = 15.9, $p < 0.001$; peak = 1700), and had a higher number of fish detected per hour than the north arm (> 0.05 fish per hour). Fish detected per hour in the north arm estuary was consistent throughout the day (< 0.002 fish per hour; Chi.sq = 1.7, $p = 0.2$). A similar pattern was observed in the lower river receivers, fish detected per hour significantly increased throughout the day in the south arm before decreasing at night (Chi.sq = 25.0, $p < 0.001$; peak = 1700), but the north arm had a higher number of fish detected per hour (> 0.1 fish per hour) consistently compared to the south arm (< 0.1 fish per hour; Chi.sq = 0.8, $p = 0.26$). Once in the mainstem of the river (river km 4), there were consistent fish detected per hour at Major Jimmy (> 0.2 fish per hour; Chi.sq = 0.0, $p = 0.96$), but a significant daily pattern observed at the fence with higher fish detected per hour observed in the morning (< 0.1 fish per hour, peak = 0500; Chi.sq = 12.7, $p < 0.001$).

Multiple environmental predictor variables had a significant relationship with depth or activity for Cowichan Chinook while in the Cowichan River. Hour of the day had a significant relationship with Chinook salmon depth ($F = 1270.3$, $p < 0.001$) and activity levels ($F = 2732.8$ $p < 0.001$), with salmon found deeper during the day and moving faster (Figure 5.7A and E).

Similarly, Julian day was also significant related to Chinook salmon depth ($F = 52.2$, $p < 0.001$) and activity levels ($F = 36.2$, $p < 0.001$), but varied through time (Figure 5.7B and F). Both temperature (depth: $F = 69.0$, $p < 0.001$; activity levels: $F = 30.5$, $p < 0.001$) and river discharge (depth: $F = 20.2$, $p < 0.001$; activity levels: $F = 7.8$, $p < 0.001$) had a significant effect on the behaviours of Cowichan Chinook in the Cowichan River, with salmon moving slower and deeper at higher temperatures and moving faster and deeper at higher river discharge (Figure 5.7C-D, G-H). Station was also significant for both depth ($F = 16710.5$, $p < 0.001$) and activity levels ($F = 1006.5$, $p < 0.001$) on behaviours in the river. Salmon were found to be deeper at the north arm station, and shallowest at the fence and north estuary locations. Salmon also tended to move fastest at the north estuary station, but moved slowest at the Major Jimmy station. Finally, fish ID had a significant effect on depth ($F = 268.1$, $p < 0.001$) and activity levels ($F = 120.4$, $p < 0.001$). Fish 8214 was generally the deepest, and 4636 was the shallowest, while fish 10618 was the fastest and 7954 was the slowest.

5.5 Discussion

Cowichan Chinook responded behaviourally to both increased sound pressure levels and the number of AIS vessels present in Cowichan Bay, British Columbia representing the first documentation of Chinook salmon responding to increased noise levels. Other species of salmon (pink and chum) were shown to have a 50% chance of responding to vessel noise when at a sound pressure level of 140 dB re 1µPa (10-20000 Hz; van der Knaap et al., 2022), but in our study we found significant changes in depth starting to occur at 120 dB re 1µPa (20-24000 Hz). Additionally, we observed that behaviours might differ based on the type of vessel and noise produced. For example, with a single AIS vessel present Chinook were at a shallower depth and more active (Figure 5.5), but at higher sound pressure levels (potentially recreation vessels that are not tracked) and with two AIS vessels Chinook were deeper with no significant change in activity. These different behaviours could be attributed to the different noise characteristics (*e.g.*, frequencies). Most (65%) of the single AIS vessel time points were from the anchored bulk carriers in the bay, which produce different noise, lower SPLs, but more constant through time (Murchy et al., 2022) compared to vessels that move through an area fast but produce higher sounds levels depending on size and speed (Picciulin et al., 2022). It is likely that at least one of the vessels in the two AIS vessel time points was moving creating different noise characteristics, changing the behaviour observed and making them similar to high SPL time points.

In other species of fish, responses to boat noise are similar to how the species responds to predators (Jacobsen et al., 2014). While Cowichan Chinook have not had their behaviour examined near predators, chum salmon that encountered Dall's porpoise (*Phocoenoides dalli*) showed a mix of responses based on the encounter's location (Yano et al., 1984). Chum salmon dove deep when they encountered the porpoise in shallow waters but remained at the surface when in deep water (> 100 m), but in both cases chum salmon remained stationary with little to no horizontal movement away (Yano et al., 1984). Additionally, resident killer whales have had their behaviour monitored during prey capture events and demonstrated deep dives, indicating salmon moving deeper to avoid predation (Tennessen et al., 2019).

The changes in the behaviours observed by Chinook salmon in this study are potentially indirectly impacting the salmon's survival and fecundity (Frid and Dill, 2002) as well as population growth (Soudijn et al., 2020), representing biologically meaningful changes. Atlantic cod (*Gadus morhua*) had reduced population growth due to acoustic impacts that increased the energy they were expending or decreased the amount of food they consumed (Soudijn et al., 2020). While the salmon were not feeding during our study, the energy they used to change their depth or increase their activity levels could have been used for reproductive output or for the migration upriver, potentially causing pre-spawning mortality or decreased fecundity. Further research would be needed to fully understand any indirect impacts from elevated noise levels on Chinook salmon.

Terminal detection and behaviour patterns of Cowichan Chinook showed clear relationships with time (hour of day and day of year), and environmental variables, demonstrating the impact these key components of the marine environment have on behaviour. Cowichan Chinook had consistent daily increases in detections in Cowichan Bay, with the largest increase in detections found along the front of the bay near the estuary, then detections falling off overnight, potentially indicating salmon moving outside the bay (not on telemetry array) overnight. Salmon also had consistent patterns in depth and activity levels, moving shallower but with less activity during the day. This shallower pattern during the day is consistent with some previous research in British Columbia (Candy and Quinn, 1999), but different from other research conducted in Cook Inlet, Alaska (Courtney et al., 2021; Welch et al., 2013). Candy and Quinn (1999) found strong diel patterns in depth distribution with salmon shallower during the day (25-64 m) compared to overnight (49 – 78 m) in Johnstone Strait BC, however, Welch et al. (2013) found Chinook salmon moved slightly shallower overnight compared to during the day in Cook Inlet Alaska, but this difference was minor (0.8 m). Courtney et al. (2021) also did not find a daily pattern in depth in Cook Inlet, but these salmon were tagged in March which could represent differences in behaviour based on environmental factors (*e.g.,* temperature, climate), Additionally, researchers used MiniPAT satellite tags that are much larger (60 g) than acoustic tags (13 g) and could have altered the salmon's behaviour compared to our study.

Tidal patterns were also found to have a relationship with behaviour in Cowichan Chinook. Our study found that Cowichan Chinook had significantly less activity during the flood tide, however, did not find a difference in depth behaviour between an ebb or flood tide. Less activity on the flood tide might be an indication of salmon milling near the estuary assessing cues (*e.g.,* olfactory, discharge rate) for river migration on the flood tide. To our knowledge, only one study has examined tidal impacts on adult Chinook salmon (Welch et al., 2013). Welch et al. (2013) examined tide directions (ebb or flood) impact on river entry and found more salmon entered the river on a flood tide. While our study did not examine the influence of tide on river entry, our finding of lower activity level on a flood tide could represent river entry on the flood tide over the ebb.

Our study was also the first to examine how environmental variables influenced the behaviours of Chinook salmon in the river. Detection patterns of Cowichan Chinook in the Cowichan estuary and river were highly dependent on accessibility. For example, in 2019 the north arm lower river was not passible until September 26 when gravel was extracted to allow fish to pass through. This obstacle to fish migration in the north arm led to more salmon migrating up the south arm in 2019. Similarly, in 2022, late-season drought conditions restricted upstream migrations and resulted in tagged salmon spending days at some locations in the river (2.25 days in North arm and 18 days at Major Jimmy: Appendix C), potentially biasing the models towards a few individuals. Additionally, behaviours in the Cowichan estuary and river were similarly restricted by water depth with the depth distribution limited by the maximum depth of each station, all stations deployed between 1 and 3 m water depth. Cowichan Chinook had higher activity levels in the river (> 1 m/s^2) compared to in the bay (~ 1 m/s^2); which is similar to Welch et al. (2013), where salmon traveled slower in the ocean and faster in the river.

Water temperatures in the river significantly affected depth and activity levels, with salmon found deeper and less active at higher temperatures (20C), depth and activity also varied with discharge level with salmon being deeper and more active at higher discharge levels.

The behaviour of tagged Cowichan Chinook immediately after tagging aligns with what has been observed in other studies (Candy et al., 1996; Candy and Quinn, 1999). Candy and Quinn (1999) recorded deep dives for the first 6 hours after tagging, followed by shallower movements. In another study, released salmon exhibited a mix of behaviours, with some salmon diving deep (300 m; vertical movement) while others remained near the surface but travelled away from the tagging site (horizontal movement; Candy et al., 1996). Additionally, different movement patterns were observed after tagging with salmon moving offshore after tagging and then moving back (Candy and Quinn, 1999), but were not evaluated in this study. Our study was also the first to examine activity (acceleration) after tagging, so these values could not be compared to previous research.

Tagging mortality found in this study appears to be comparable to other studies tagging adult salmon (Bendock and Alexandersdottir, 1993; Candy et al., 1996). Post-tagging mortality rates of Chinook salmon captured in a purse seine were between 0-36% depending on the tagging year, with overall mortality 23% for all years combined (Candy et al., 1996). Candy et al. (1996) also found the biggest indicator of survival was a shorter landing time (mean = 22 minutes). Salmon captured in-river using recreational angling gear had shorter landing times (< 10 minutes) and subsequently better survival compared to purse seine captures, with mortality ranging from 4.1-10.6% (Bendock and Alexandersdottir, 1993). Our study also had shorter landing times (< 10 minutes), resulting in a similar mortality of 10.6% for all combined years.

This study also recorded some predation events, similar to other research (Courtney et al., 2021). One salmon tagged in 2020 had a significant change in behaviour recorded approximately five hours after tagging with activity levels increasing to above 2 m/s^2, and depths being deeper on average. This change likely indicates a predation event where the acoustic tag was consumed, and the seal's behaviour was recorded for two days before it was detected on the bottom of the bay. A seal likely consumed another Cowichan Chinook in the lower river in 2019. Activity levels maxed out (4.9 m/s^2) multiple times while the salmon did repeated dives to the bottom of the river (~ 3 m), indicating a potential chase. The tag abruptly stopped moving (0 m/s^2) and remained at a consistent depth (0.7 m); this salmon was likely consumed, but the tag fell out during consumption and was not consumed by the predator. The last Cowichan Chinook in 2019, which disappeared in the estuary, could have been consumed by a seal but no tag data were available, so this cannot be confirmed. Additionally, two Cowichan Chinook in 2019 were last detected in the south arm of the lower river. There are two bridges between the last detection and the next receiver, where members of Cowichan Tribes fish for adult salmon. So, it is probable that these two salmon were captured as a part of Cowichan Tribes subsistence fishery.

The predation pressure from pinnipeds likely influenced the behaviour patterns of Chinook in Cowichan Bay. Cowichan Bay has a high presence of Pacific harbour seals (*Phoca vitulina*) during August and September when the adult salmon return to spawn. Harbour seals are nocturnal feeders (Frost et al., 2001; Wilson et al., 2014) and tend to forage within 30 km of their haul-outs (Tollit et al., 1998). The daily patterns in movement and behaviours could thus be indicative of an antipredator behaviour. Cowichan Chinook tagged in this study showed movement out of the bay at night away from the primary haul-out for harbour seals (estuary log booms), while also showing strong diel patterns in depth (deeper) and activity (more activity) at night when more seals are likely foraging. Previous work on adult Chinook salmon depth patterns did not show strong diel movements (Courtney et al., 2021), and Welch et al. (2013) observed the opposite with salmon being found shallower at night, potentially indicating predation pressure is driving the response observed in Cowichan Bay. While Cook Inlet is important habitat for pinnipeds (Montgomery et al., 2007) and predation events have been observed (Courtney et al., 2021), the size of Cook Inlet (290 km long) is much larger than Cowichan Bay (4 km long), allowing for more haul out sites and less overlap between pinnipeds and salmon. In Cowichan Bay, salmon might need to employ both spatial and behavioural antipredator responses due to the high concentration of pinnipeds near the estuary. Additionally, as Cowichan Chinook approached the estuary their activity levels were higher compared to at the opening to the bay, and had higher activity levels at low tide; both of these increases in activity suggest the potential for more burst behaviours (Murchie et al., 2011) as they are likely encountering pinnipeds in the water near their haul-outs.

Significant relationships between daily, environmental, and acoustic variables were found in this study, highlighting the importance of these marine ecosystem components on Cowichan Chinook behaviour and movements; however, this study is not without limitations. One main limitation was that the sound pressure levels were not calculated as received levels at the individual fish, but at the station location. Depending on the location of the noise source and the fish within the 1 km detection radius (estimated detection range of tag), the sound pressure levels were likely higher or lower at the fish compared to the underwater hydrophone. Additionally, the particle motion component of the sound sources was not accounted for. Chinook salmon are hearing non-specialists with the best hearing between 100-300 Hz and are more sensitive to sound sources' particle motion component (Hawkins and Johnstone, 1978; Oxman et al., 2007).

Including received sound levels at the fish and particle motion were not possible during this study due to technological limitations (Miksis-Olds et al., 2018), but should be considered in future work (*e.g.,* controlled studies, technological advances). Lastly, movement and behaviour patterns in Cowichan Chinook could only be evaluated on a coarse scale and individual fish could not be tracked for more fine-scale movements within Cowichan Bay. Future work should deploy a smaller grid of acoustic receivers or use active tracking of individual salmon to understand the patterns found in this study on a finer scale. Tracking salmon movements and behaviour on a finer scale would allow for more interactions with vessels to be tracked, like closest point of approach to get distances, and fine scale movement patterns, to understand smaller movements that occurred within the range of a single receiver (1 km) in this chapter.

Chinook salmon are an important component of the Northeast Pacific Ocean, representing an ecologically, economically, and culturally significant keystone species. Understanding adult movement patterns prior to river spawning migrations and the impacts of underwater noise represent two areas of research that are vital to aiding in recovering these declining populations (Riddell et al., 2013; Welch et al., 2021). Our results demonstrated the influence of hour of the day on detection and behaviour patterns in Cowichan Chinook, and that tide height influenced depth and activity levels. We also found significant changes in behaviours from increased sound levels and the number of AIS-equipped vessels present, the first study to show direct impacts on the behaviour of Chinook salmon. Underwater noise levels have been increasing due to anthropogenic sources and have been shown to impact resident killer whales (Williams et al., 2019, 2002), Pacific herring (van der Knaap et al., 2022), and now Chinook salmon demonstrating ecosystem-wide changes to sensitive populations. Underwater noise levels are likely to continue rising with increased vessel production to meet trade requirements, and understanding the connections between the responses of each of these species will be required to protect these important species.

Table 5. 1. Equipment moorings in Cowichan Bay.

Location	Latitude (N)	Longitude (W)	SoundTrap (Y/N) (Years)	Water depth (m)	Receiver depth (m)
N 2A	48.7589	123.612	Y (2019/2020)	19	18
Front Mid Bay	48.7523	123.619	Y (2019/2020/2022)	45	44
Marina	48.7455	123.622	Y (2019/2020)	23	3
Genoa Bay	48.7556	123.598	Y (2019/2020/2022)	19	18
Mid Bay	48.7502	123.609	Y (2022)	52	51
S Mid Bay	48.7394	123.605	Y (2019/2020)	22	20
N Bay Opening	48.7450	123.575	Y (2019/2020)	18	17
Mid Bay Opening	48.7415	123.582	N	63	62
S Bay Opening	48.7334	123.583	Y (2019/2020)	15	13
Sep Point 1	48.7456	123.568	Y (2020)	25	24
Sep Point 2	48.7509	123.567	N	104	103
N Estuary	48.7695	123.635	N	2.3	2
S Estuary	48.7557	123.64	N	2.4	2.1
N Lower River	48.7664	123.645	N	1.5	1.2
S Lower River	48.7603	123.651	N	3	2.7
Major Jimmy	48.7732	123.665	N	2.4	2.1
Fence	48.7719	123.713	N	1.2	0.9

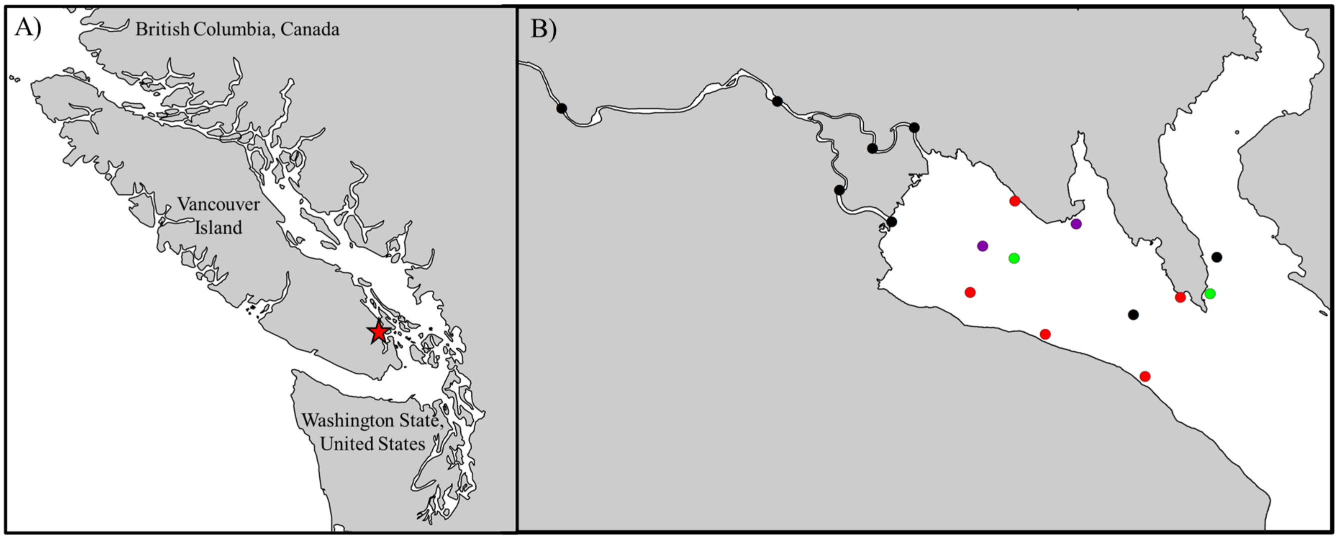

Figure 5. 1. A. Map of Vancouver Island with portions of lower British Columbia and western Washington State. Red star denotes Cowichan Bay. B: Close up map of Cowichan Bay, with acoustic tag receiver locations denoted with black dots and acoustic tag receivers paired with SoundTrap hydrophones denoted with coloured dots based on the number of years a SoundTrap was deployed there (Green = 1 year, Red = 2 years, Purple = 3 years). BCMap obtained from Natural Earth and BC Provincial data catalog.

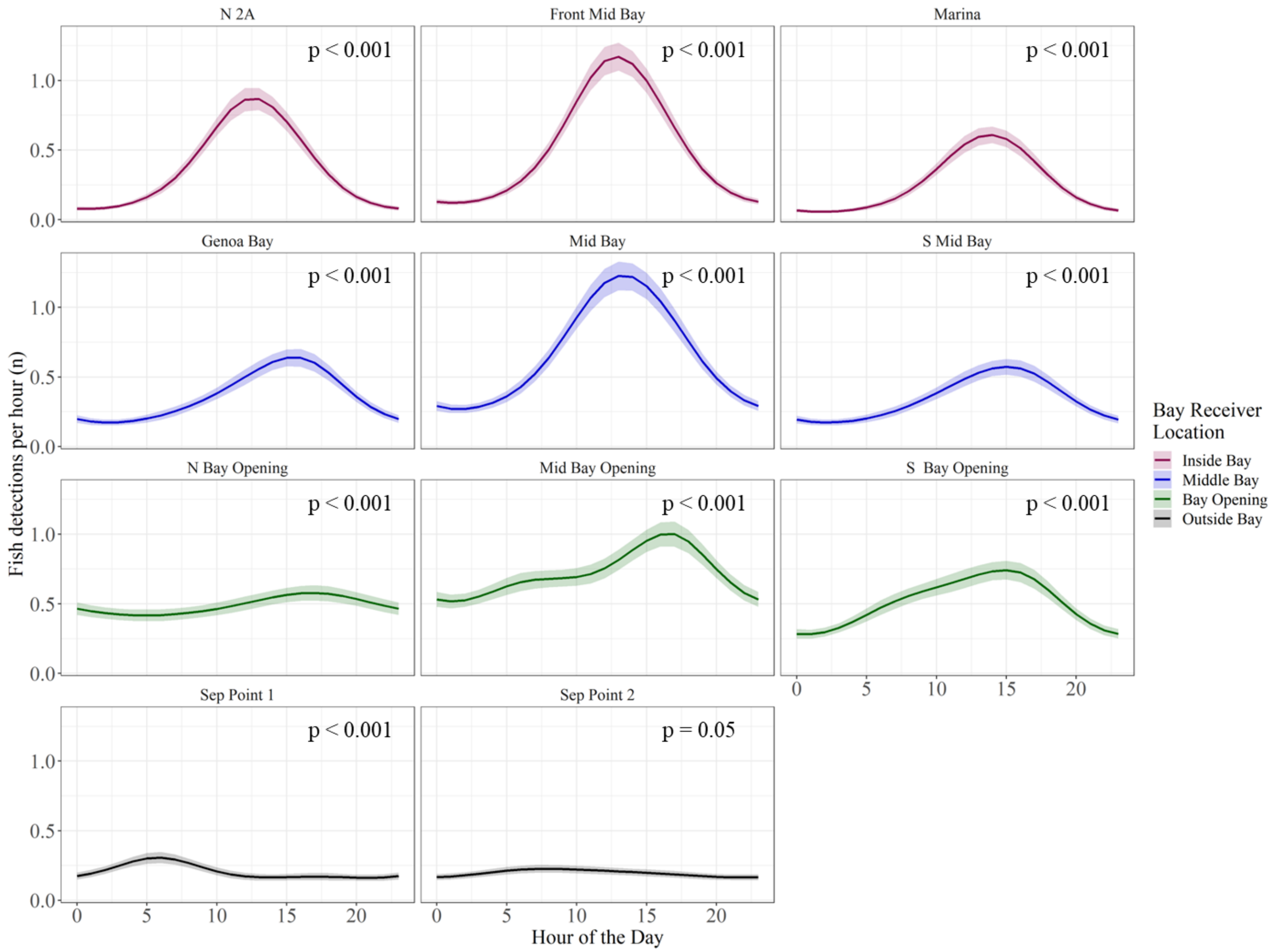

Figure 5. 2. Model predicted detections by hour of the day for Cowichan Chinook at each station in Cowichan Bay, colours denote general location within the bay.

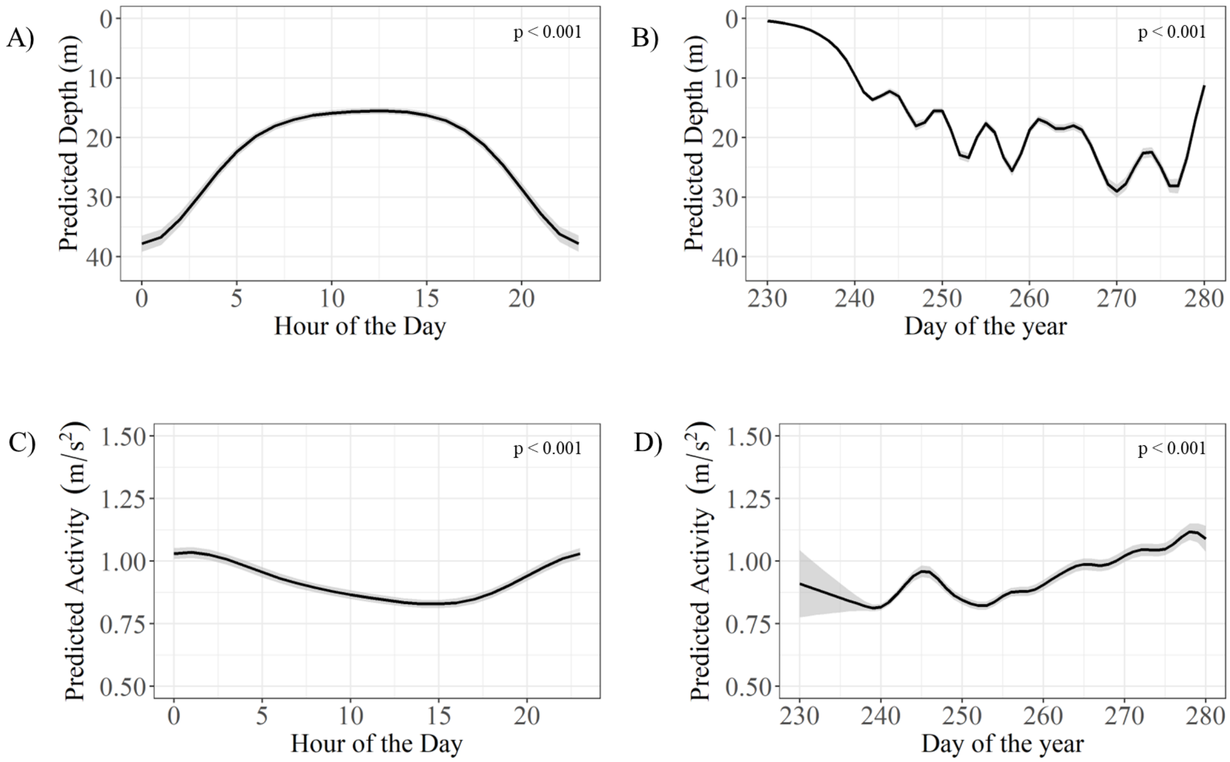

Figure 5. 3. Model predicted behaviours for Cowichan Chinook. A) Predicted depth by hour of the day. B) Predicted depth by day of the year. C) Predicted activity levels by hour of the day. D) Predicted activity levels by day of the year.

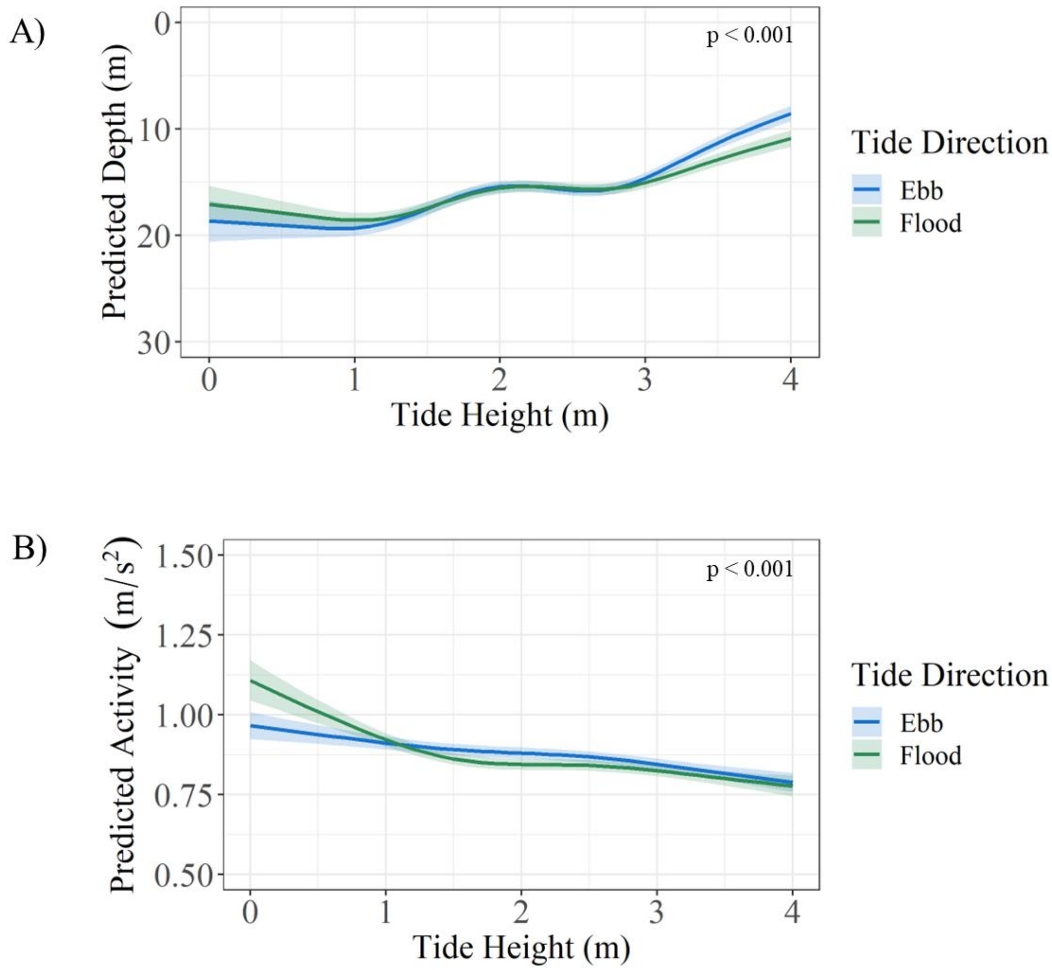

Figure 5. 4. Model predicted behaviours for Cowichan Chinook. A) Predicted depth by tide height. B) Predicted activity levels by tide height.

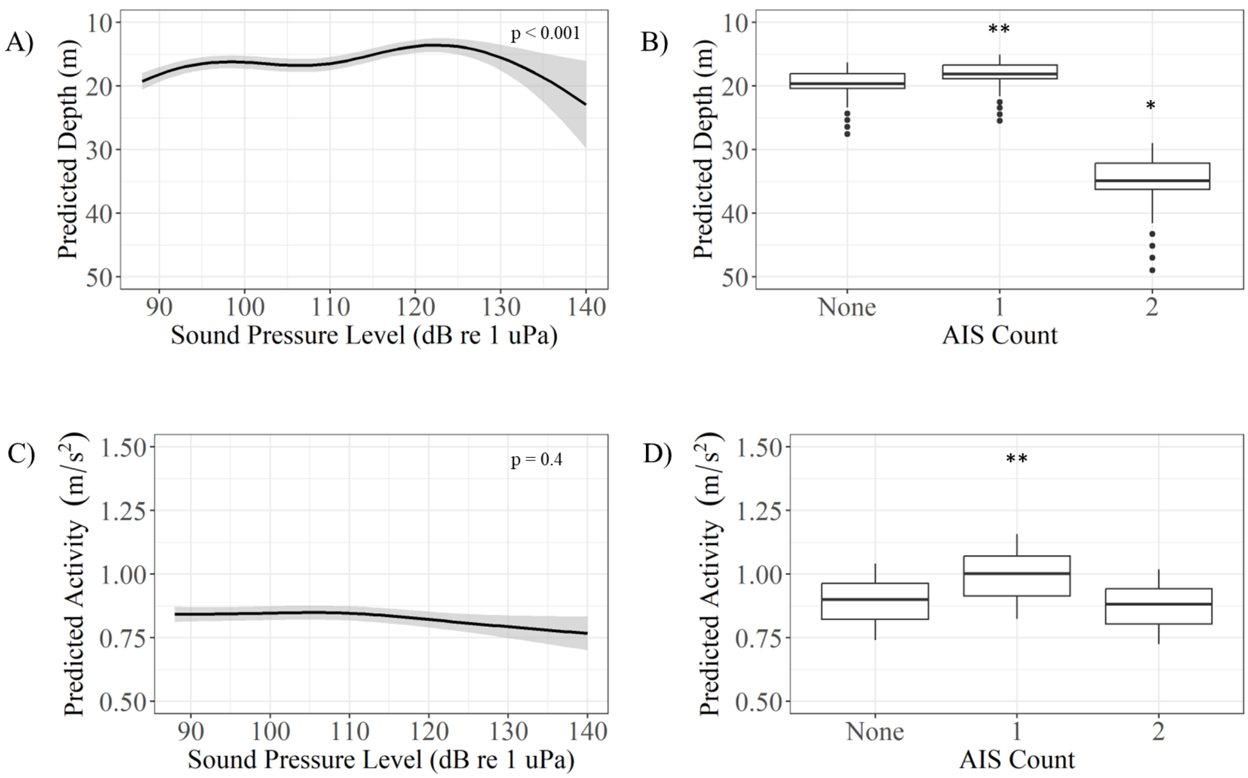

Figure 5. 5. Model predicted behaviours for Cowichan Chinook. A) Predicted depth by sound pressure level (dB re 1 μPa). B) Predicted depth by the number of AIS vessels. C) Predicted activity levels by sound pressure level (dB re 1 μPa). D) Predicted activity levels by the number of AIS vessels. Asterisks (**) indicate significantly different predicted depth and activity levels at p < 0.05 and a single asterisk (*) indicates significance at p = 0.05.

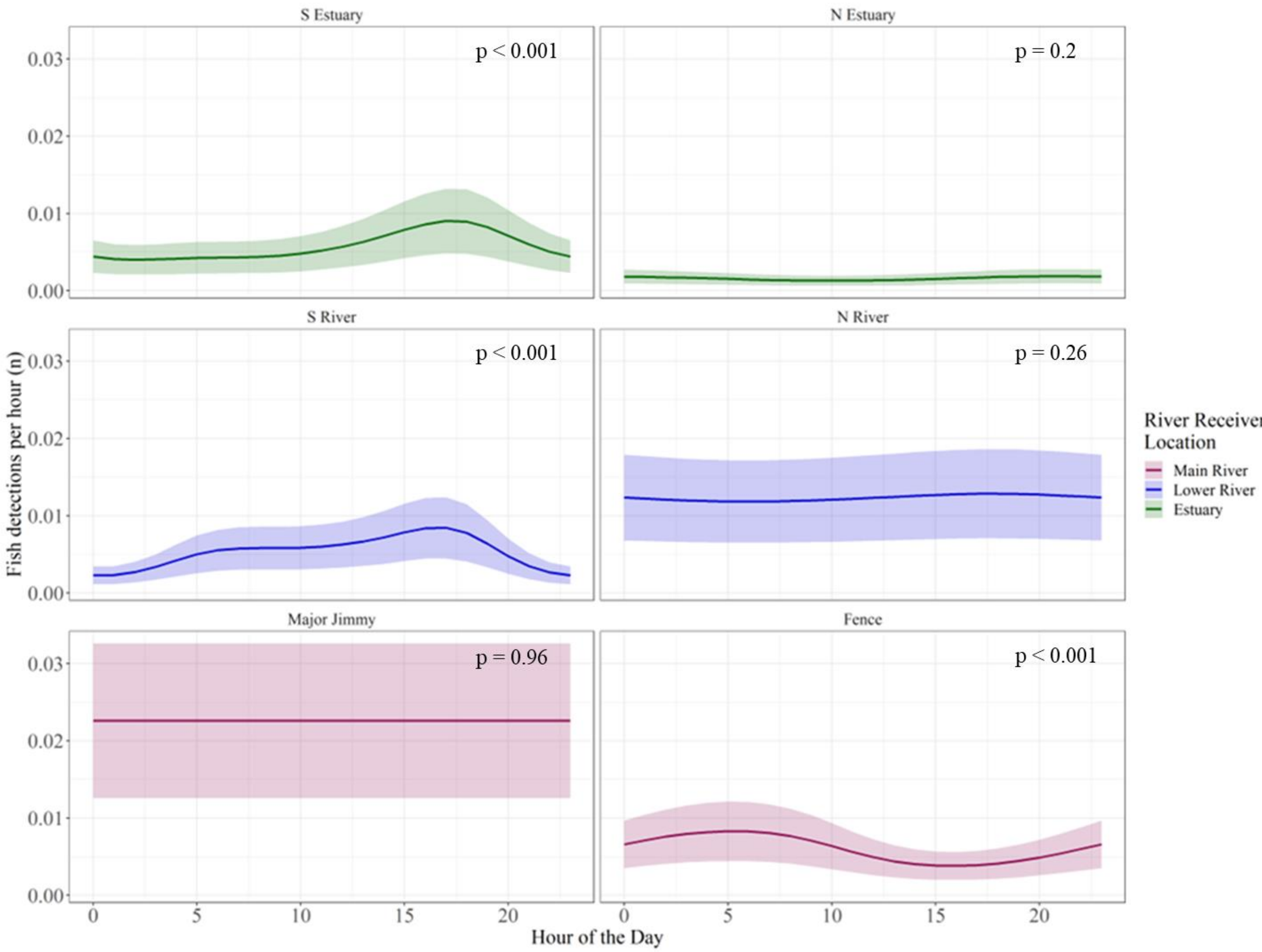

Figure 5. 6. Model predicted detections by hour of the day for Cowichan Chinook at each station in Cowichan estuary and river, colours denote general location within the river.

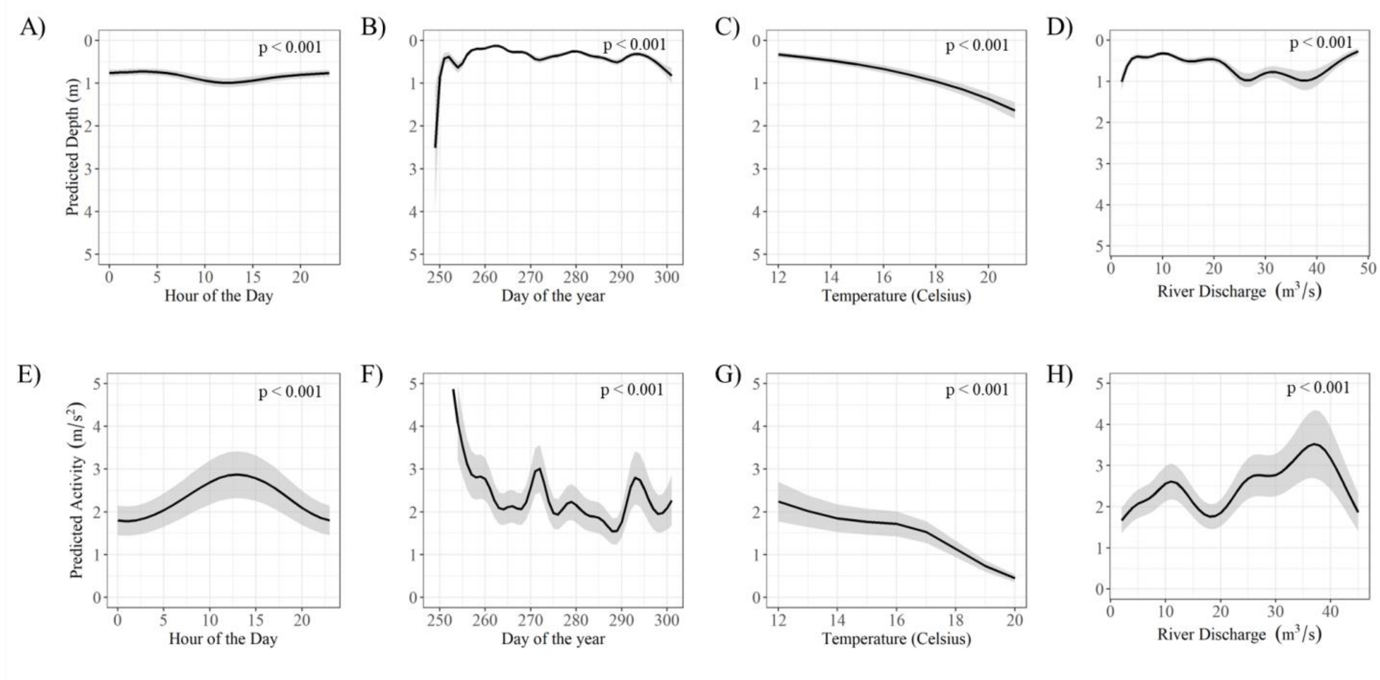

Figure 5. 7. Model predicted behaviours for Cowichan Chinook in the estuary and river. Predicted depth for A) Hour of the day. B) Day of the year. C) Temperature (°C). D) River discharge. Predicted activity levels for A) Hour of the day. B) Day of the year. C) Temperature (°C). D) River discharge.

Chapter 6 – Conclusion

The underwater soundscape around the world has been changing (McDonald et al., 2006) with the continued growth of the shipping industry (Hildebrand, 2009; Tournadre, 2014), which dominates low frequency noise (40- 1000 Hz) leading to ambient underwater noise levels increasing by 10-12 dB in some areas (Ainslie et al., 2021; Ross, 1976). Locally, in the waters around southern British Columbia and Washington State (Salish Sea), there are high amounts of commercial vessels that pass through daily headed to the Ports of Vancouver and Seattle (Veirs et al., 2016), creating hotspots of noise in Haro Strait, Juan de Fuca and Johnstone Strait (Erbe et al., 2014). The Port of Vancouver is considering an expansion of one of the terminals (Terminal 2, Roberts Bank), which is expected to double the capacity of the port, if approved, (Vancouver Fraser Port Authority, 2018) increasing the number of vessels travelling through the Salish Sea and potentially anchoring in its coastal ecosystems. A better understanding of the impacts of vessel noise on the marine soundscape and the marine species that inhabit it represents a key conservation concern.

6.1 Soundscape changes from underwater noise

Noise from vessels in motion has been well described in the literature (*e.g.*, Arveson and Vendittis, 2000; Gassmann et al., 2017; Halliday et al., 2020; McKenna et al., 2012; Wittekind and Schuster, 2016), and has shown to significantly alter the underwater soundscape (Burnham et al., 2021; Pine et al., 2021). In recent years, mitigation strategies have been used to reduce the amount of noise produced by vessels (Williams et al., 2019). For example, a voluntary vessel slowdown initiative run by the Enhancing Cetacean Habitat and Observation (ECHO) program at the Vancouver Fraser Port Authority decreased underwater noise levels produced by large commercial by 2.7 dB in Haro Strait and 2.8 dB in Boundary Pass between June and October

2022, when compared to pre-slowdown periods (Vancouver Fraser Port Authority, 2023). Similarly, commercial vessels that changed their speed to around 11 knots reduced sound pressure levels by 6-11 dB depending on the type of vessel (MacGillivray et al., 2019), with median reductions in sound pressure levels highest for between 10-100 Hz (3 dB) and lowest for between 10,000-100,000 Hz (0.3 dB: Joy et al., 2019). Other potential mitigation methods to reduce sound pressure levels include modifying ship design (Spence and Fischer, 2017) or moving ship lanes (Williams et al., 2019). However, mitigation methods have been mainly focused on large commercial vessels while in motion.

This dissertation has presented evidence that bulk carriers also emit noise and raise broadband sound pressure levels (20-24000 Hz) by 2 to 8 dB re 1 μPa when carriers are anchored. As a result, the presence of anchored vessels adds noise to the acoustic ranges of many marine species, potentially masking conspecific signals, altering daily diel patterns in sound pressure levels, and reducing the time at lower SPL necessary for marine species to detect biologically relevant sounds. Similarly, significant changes to the underwater soundscape were also observed from a smaller commercial vessel (tugboats). Tugboats significantly raised the underwater noise levels by 6 to 10 dB re 1 μPa (20-24000 Hz), showing another anthropogenic noise source that might alter behaviour in marine species and that is currently not being considered for mitigation.

6.2 Chinook salmon behaviour

Examination of behaviours (acoustic and movements) of adult Chinook salmon (*Oncorhynchus tshawytscha*) has been minimally explored in the literature (Candy and Quinn, 1999; Courtney et al., 2021; Neproshin, 1972; Welch et al., 2013). The first research to examine sound production in Chinook salmon occurred in the 1970s but provided limited information on acoustic properties (*e.g.*, frequency range, durations) and was more anecdotal (Neproshin, 1972). Neproshin (1972) did document potential air movement sounds in Chinook salmon, with whistle sounds (frequencies up to 6000 Hz), low frequency knocks (frequency range: 100-500 Hz), and rumbling (frequency range: 300-500 Hz) sounds observed. In chapter 4, Chinook salmon were observed to produce multiple types of air movement sounds, including a gill-bubble FRT and a moan sound that have been recorded in other salmonids (Rountree et al., 2018). Chinook salmon were also documented to produce hydrodynamic sounds at the surface and a pulse sound, but the mechanism to produce the pulse sound could not be identified. These sounds were found to be different from other Pacific salmon (coho salmon, *Oncorhynchus kisutch*, pink salmon *Oncorhynchus gorbuscha*) in frequency and duration parameters, indicating the potential for passive acoustic monitoring.

Ocean movement and behaviour patterns of adult Chinook salmon have also received little research attention, but overall patterns have emerged (Candy and Quinn, 1999; Courtney et al., 2021; Welch et al., 2013). Chinook salmon that are close to their natal rivers tend to be closer to the surface, spending more time at depths of less than 10 meters (Courtney et al., 2021; Welch et al., 2013), and traveling close to shore staying within 10 kilometers (Candy and Quinn, 1999; Welch et al., 2013). Daily patterns in depth distribution have also been documented but with different patterns observed in different areas. In Cook Inlet, Alaska, little to no depth patterns were observed, with slightly deeper depths observed during the day (Courtney et al., 2021; Welch et al., 2013). However, in Johnstone Strait, British Columbia Chinook salmon were shallower during the day (Candy and Quinn, 1999), demonstrating the opposite pattern. In Chapter 5, we also observed a strong daily pattern in depth distribution, with Chinook salmon found shallower during the day and moving deeper overnight. Daily patterns in activity levels, with Chinook salmon less active during the day and more active at night, and detection patterns, increased detections of tagged salmon in the bay during the day and decreasing overnight, were also observed. These represent the first indication of multiple daily movement and behaviour (depth and activity) patterns prior to river migration.

Environmental variables are key components of marine ecosystems, but their inclusion in research on movements and behaviour in salmon has been negligeable. Welch et al. (2013) found more salmon moved into the river during a flood tide compared to an ebb tide, but the height of the tide was not included. Our research found that tide height was important in predicting depth and activity levels, with salmon found shallower and less active at high tide. Tide direction (flood or ebb) was only found to be significant on activity levels, with salmon moving less on a flood tide potentially indicating movement into the river similar to what Welch et al. (2013) documented.

6.3 Ecosystem consequences from underwater noise

Even though research into anthropogenic noise and impacts to marine species has been increasing since the 1970s (Duarte et al., 2021), little research has been conducted on how changing underwater noise conditions could alter predator-prey dynamics. As an example, resident killer whales (*Orcinus orca*) have been extensively studied to examine the impacts of vessels on their behaviours (*e.g.*, Lacy et al., 2017; Williams et al., 2009, 2002), but only recently is more research being conducted to understand the impacts of vessels on their foraging (Lusseau et al., 2009). When an increased number of vessels are found within close proximity (<400m) of resident killer whales, resident killer whales have been documented to change their behaviour from feeding to traveling (Lusseau et al., 2009). This behavioural shift indicates a potential displacement of killer whales from their optimal foraging areas. Vessels are also documented to increase dive times and decrease the probability of prey capture in tagged resident killer whales (Holt et al., 2021). Additionally, sounds produced from passing ships elevate the frequencies (20-80 kHz) used by resident killer whales for echolocation by 12-17 dB re. 1μPa (Veirs et al., 2016), and could alter their foraging efficiency (Burnham et al., 2023; Holt et al., 2021). Acoustic models of shipping noise have found that the range for communication frequencies (1-40 kHz) are reduced and likely do not exceed ambient levels, reducing the ability of whales to maintain social contact. Similarly, echolocation frequencies (50 kHz) were most impacted in shipping lanes and at 10 m depth, where echolocation signals would not be heard above ambient levels, impacting the whales' ability to locate prey (Burnham et al., 2023).

Examination of impacts to foraging and antipredator behaviour has also been examined in several fish species (Simpson et al., 2016, 2015; Voellmy et al., 2014a, 2014b). Exposure to noise led to the three-spined stickleback (*Gasterosteus aculeatus*) making more unsuccessful strikes on *Daphnia*, and stickleback had reduced consumption (Voellmy et al., 2014a). Similarly, the European minnow (*Phoxinus phoxinus*) experienced decreased feeding in the presence of the same noise source, but the number of unsuccessful strikes did not change significantly (Voellmy et al., 2014a). Noise disturbances were also associated with increased mortality due to predation, as European eels (*Anguilla anguilla*) displayed a reduced likelihood of startling in response to a simulated predator when exposed to noise. Additionally, when startled, their responses were slower, and they were caught more rapidly compared to controls (Simpson et al., 2015). Likewise on a reef, boat noise playback significantly reduced the survival of Ambon damselfish (*Pomacentrus amboinensis*), which was potentially facilitated by a reduction in startle responses (Simpson et al., 2016).

While not examined for antipredator behaviours, Pacific herring (*Clupea pallasii*) demonstrate a negative response to vessel noise (Schwarz and Greer, 1984; van der Knaap et al., 2022). Pacific herring have been observed to stop feeding, become a tighter school and move deeper in the water column as well as horizontally away from vessel noise playback (Schwarz and Greer, 1984). Behavioural response, including school cohesion, school orientation and swimming speed, in response to boats has also been evaluated, with changes in behaviour occurring at 123 dB re 1 µPa (10-20000 Hz; van der Knaap et al., 2022). Atlantic herring (*Clupea harengus* L.) have also been observed to respond to vessels by consistently diving to deeper depths, demonstrating avoidance (Skaret et al., 2006).

While no research to date has been conducted on the impacts of vessel noise on Chinook salmon, other species of salmon have been examined. Juvenile pink and chum (*Oncorhynchus keta*) were shown to behaviourally respond to vessels at sound pressure levels of 140 dB re 1 µPa (10-20000 Hz; van der Knaap et al., 2022), and move away from noise produced from pile driving (Feist et al., 1992). Juvenile Chinook salmon have been shown to produce a startle response and avoidance to a 10 Hz tone (Knudsen et al., 1997). The work found here in my dissertation shows the first example of adult Chinook salmon responding to noise produced by vessels. My results demonstrated that Chinook salmon were found significantly deeper at 140 dB re 1µPa (20-24000 Hz), but no significant difference was observed for activity levels. Additionally, significant changes in behaviour were observed for different number of vessels, with a single AIS-equipped vessel resulting in significantly shallower depth and significantly more activity, where two were significantly deeper (Figure 5.5).

Taken all together, these changes in behaviours across a food chain (herring, salmon, killer whales), suggest ecosystem-wide consequences from increased sound pressure levels and number of vessels. Ceasing to feed in the presence of vessel noise has been observed in resident killer whales (Lusseau et al., 2009) and Pacific herring (Schwarz and Greer, 1984), while changes in behaviour has been observed in all three species (van der Knaap et al., 2022; Williams et al., 2002). Understanding the potential impact that increased vessel noise has on these species and their predator-prey interactions would be an important step in better understanding the full extent of the influence of underwater noise. Similarly, population level assessments have not been evaluated for any of these species. In other fish and marine mammals, non-lethal individual impacts due to increased noise levels have generated significant population level consequences, including reduced growth and decreased fecundity (Pirotta et al., 2018; Soudijn et al., 2020), which would likely be observed in all three of these vulnerable species.

6.4 Future directions

The research conducted in this dissertation represents a starting point for continued examination of the impacts of shipping noise on the marine soundscape generally and Chinook salmon behaviour more specifically. First, the ongoing investigation into the effects of anchored commercial vessels is a significant conservation issue. These vessels can be anchored for extended periods, causing significant changes in the underwater soundscape over time (Murchy et al., 2022). Data presented in chapter 2 were only collected over a short time period and were not evaluated for the potential impacts to marine species. These vessel anchorages overlap with critical habitat for many marine species, but the impacts of the elevated noise levels produced from anchored vessels has yet to be evaluated on marine species. Additionally, seasonal changes in oceanic conditions have been observed to impact the noise generated by commercial vessels in motion (McKenna et al., 2013), where lower received levels are found in summer or fall because the warmer water can trap sound waves at the surface reducing transmission (Jensen et al., 2011). Similar seasonal trends likely occur with noise from anchored vessels.

Another area that needs more research is the potential impact or disruption in the sounds produced by Chinook salmon from increased noise levels. Other species of fish have been documented to increase the amplitude of the sounds they produce or have change to the fundamental frequency (Brown et al., 2021; Holt and Johnston, 2014; Luczkovich et al., 2016) to compensate for increased noise levels. The reason salmon produce sounds is unknown, the sounds may be for communication or purely incidental, but if sounds are used for communication understanding the impact of vessel noise on their sounds is a vital step in protecting this important marine species.

Finally, more research is needed to understand the influence of vessels on Chinook salmon behaviour. Results found in chapter 5 of this dissertation represent a starting point in examining how vessel noise from different types of vessels might affect salmon behaviour and movements. My results demonstrated a difference in the behaviours between high sound pressure levels (deeper depths) and a single AIS-equipped vessel (shallower depths), but when two vessels were present a similar pattern (deeper depths) was observed. While for activity, high sound pressure levels and two AIS vessels were not shown to be significant, one AIS vessel had a significant effect on activity with higher activity levels detected. The differences in behaviours could be attributed to the noise characteristics (*e.g.,* sound levels, frequencies) and differences observed between vessel types and whether they were moving or anchored. Additionally, spatial changes with increased noise levels were not accounted for in our analysis. Movement away from vessels has been demonstrated in other fish species (Ivanova et al., 2020), and salmon could be potentially avoiding areas of higher sound levels. Understanding the impact of these different vessel types on behaviours and movement patterns would greatly increase the knowledge on how salmon are being affected by vessel noise and how these changes might affect their survival.

6.5 Conclusions

Increased sound levels due to human generated noise significantly affects marine species across a variety of taxa, with growing research on plants (Solé et al., 2021), invertebrates (Murchy et al., 2020; Putland et al., 2023), fish (Cox et al., 2018; Mills et al., 2020), and marine mammals (Erbe et al., 2019; Martin et al., 2023). Currently, Canada does not have an official management strategy or policies in place to regulate underwater noise on their coasts. The research included in this dissertation fills some of the gaps in understanding the impacts of shipping noise on the marine soundscape and a key species in the Pacific Ocean, Chinook salmon. Significant changes to the underwater soundscape were documented from anchoring bulk carriers and tugboats, and changes in behaviour of adult Chinook salmon were observed in response to increased noise levels and number of AIS vessels, representing the first study on the impacts of vessels on Chinook salmon. With noise levels likely to continue to increase due to human activities, understanding how marine species might be affected both individually and collectively through population consequences or predator/prey interactions represents a significant field of study needed to protect ecologically, and culturally important marine species.

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Appendices

Appendix A: Supplemental Figures for Chapter 2

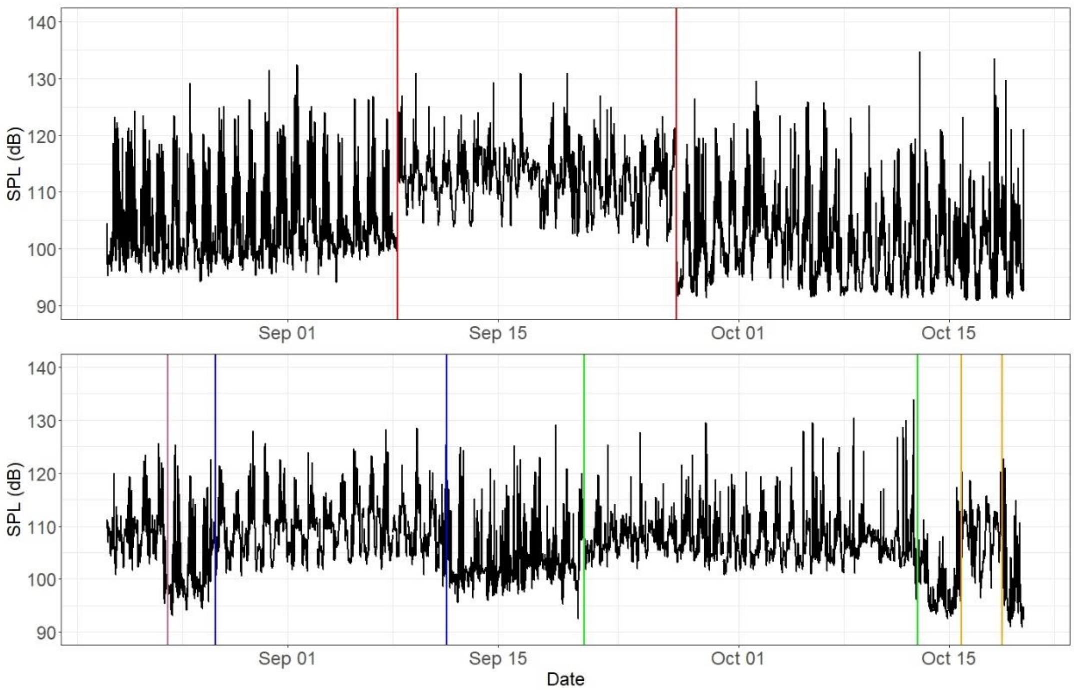

Figure A2.1: Time series of sound pressure level for 20-24000 Hz at 700 m from anchorage location. A) 2019 with red lines representing arrival and departure of anchored bulk carrier (Vessel 1). B) 2020 with lines representing arrival and departure of anchored carriers; Vessel 2 (pink), Vessel 3 (blue), Vessel 4 (green), and Vessel 5 (orange).

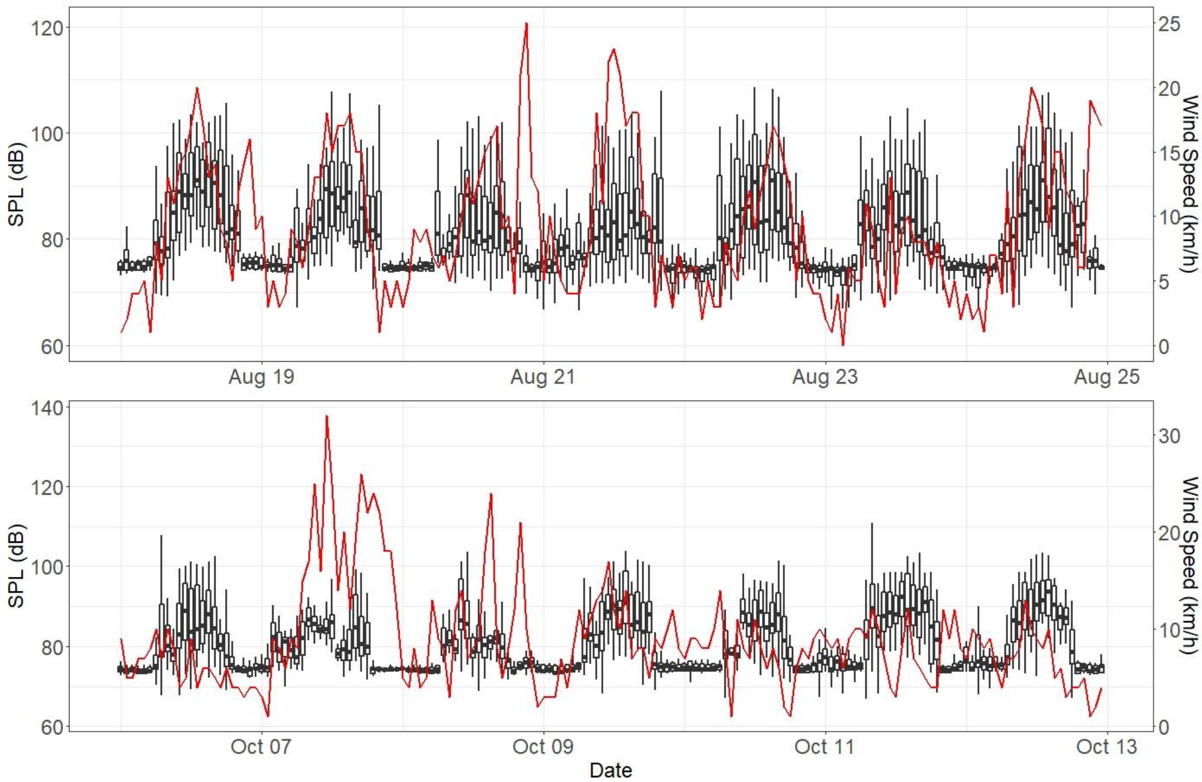

Figure A2.2: Boxplots representing the diel pattern of sound pressure levels in the 7500-8500 Hz range for all stations in Cowichan Bay for one week in August and October 2019. Red line represents wind speed data during corresponding week in August and October 2019.

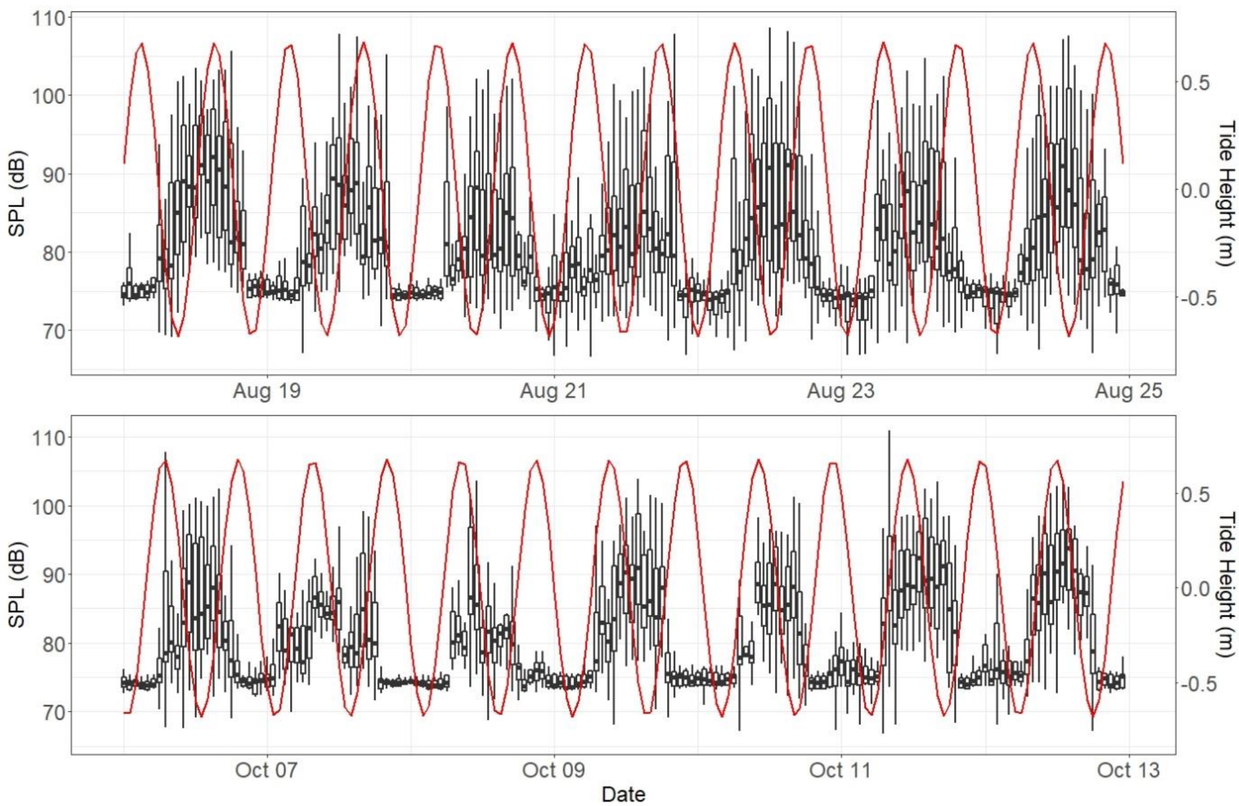

Figure A2.3: Boxplots representing the diel pattern of sound pressure levels in the 7500-8500 Hz range for all stations in Cowichan Bay for one week in August and October 2019. Red line represents tide height data during corresponding week in August and October 2019.

Appendix B: Supplemental Figures for Chapter 4

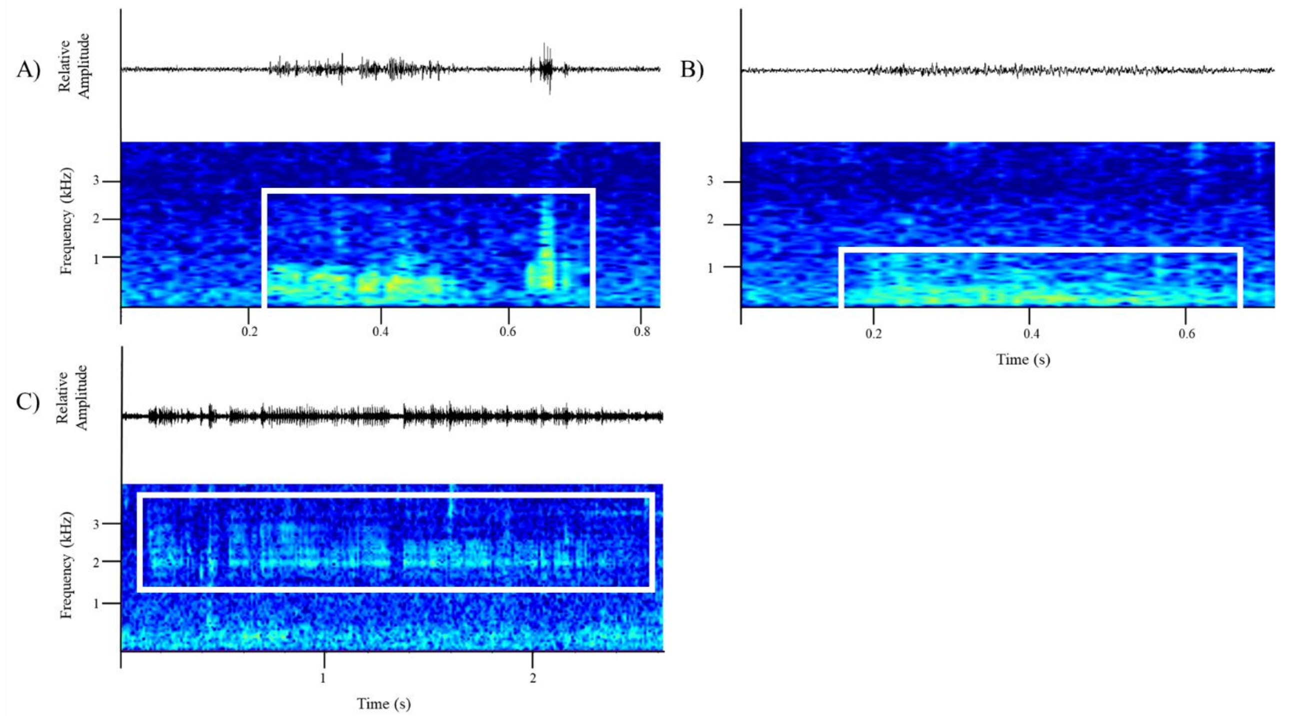

Figure B4.1: Examples of sounds produced by Chinook salmon. Top panel: the waveform filtered to each sound and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). A) Gurgle (SuppPub3) B) Growl (SuppPub4) C) Buzz. (SuppPub5)

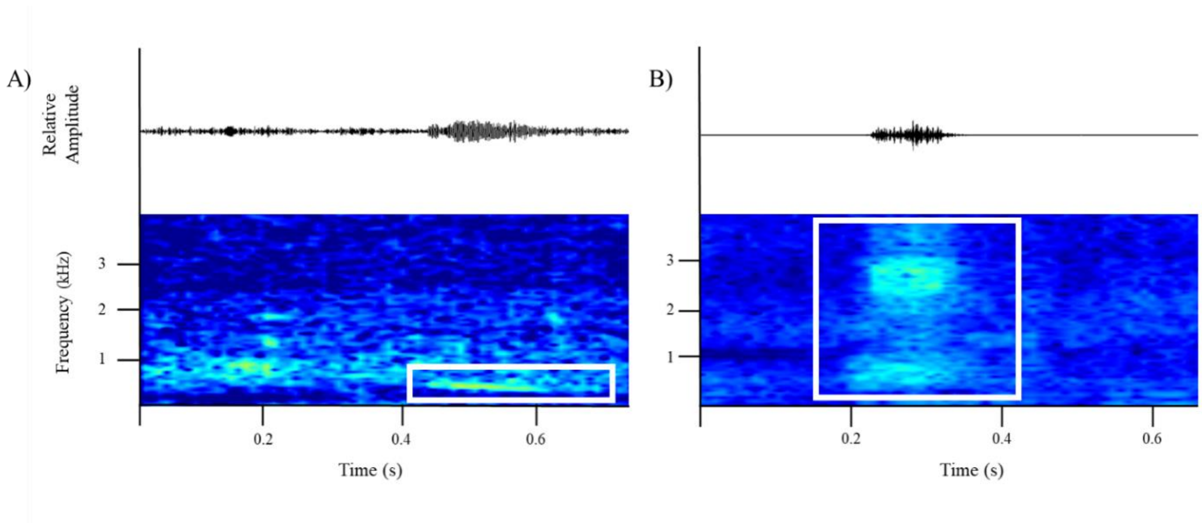

Figure B4.2: Examples of sounds produced by Coho salmon. Top panel: the waveform filtered to each sound and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). A) Moan (SuppPub6) B) vFRT (SuppPub7)

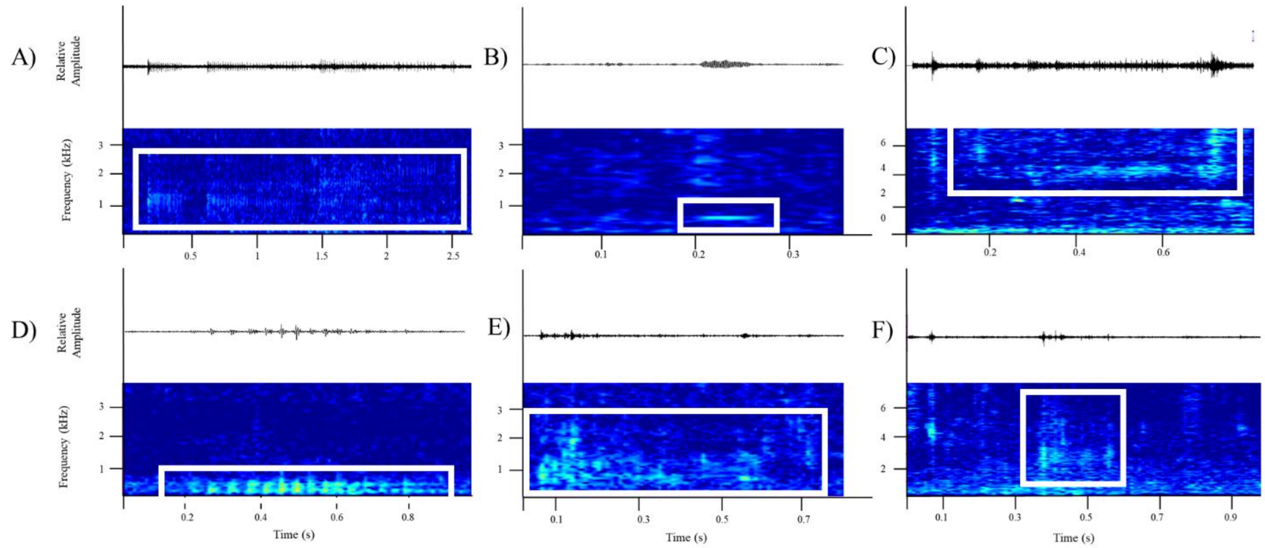

Figure B4.3: Examples of sounds produced by mixed schools of pink and Chinook salmon. Top panel: the waveform filtered to each sound and bottom panel: the spectrogram (unfiltered, 1,024 FFT, Hanning window, 50% overlap). To accommodate varying frequency ranges of sounds, subplots have different frequency ranges. A) FRT (SuppPub8) B) Moan (SuppPub9) C) Buzz (SuppPub10) D) Growl (SuppPub11) E) Gurgle (SuppPub12) F) Raspy. (SuppPub13)

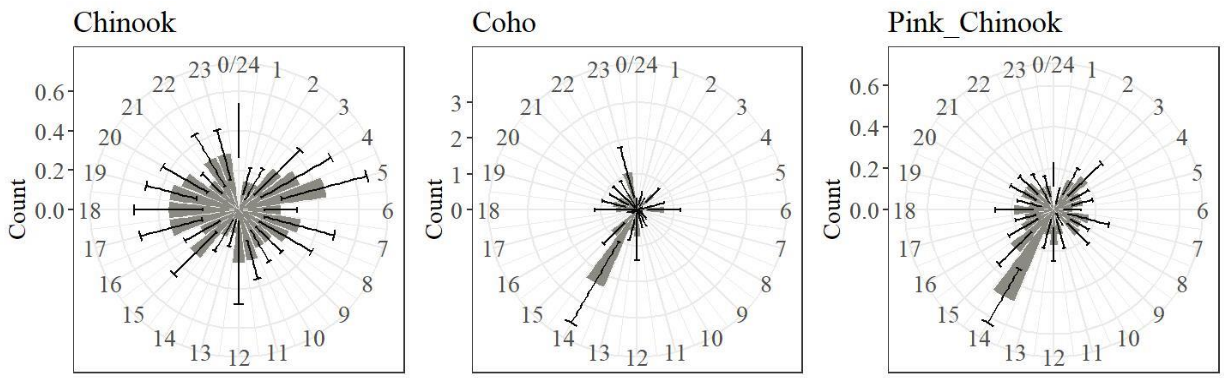

Figure B4.4: Circular bar graphs of mean and standard error of counts of all sounds produced by hour of the day in each grouping of salmon, Chinook only ($n_1 = 8$), Coho salmon ($n_1 = 2$), Pink and Chinook mixed ($n_1 = 7$).

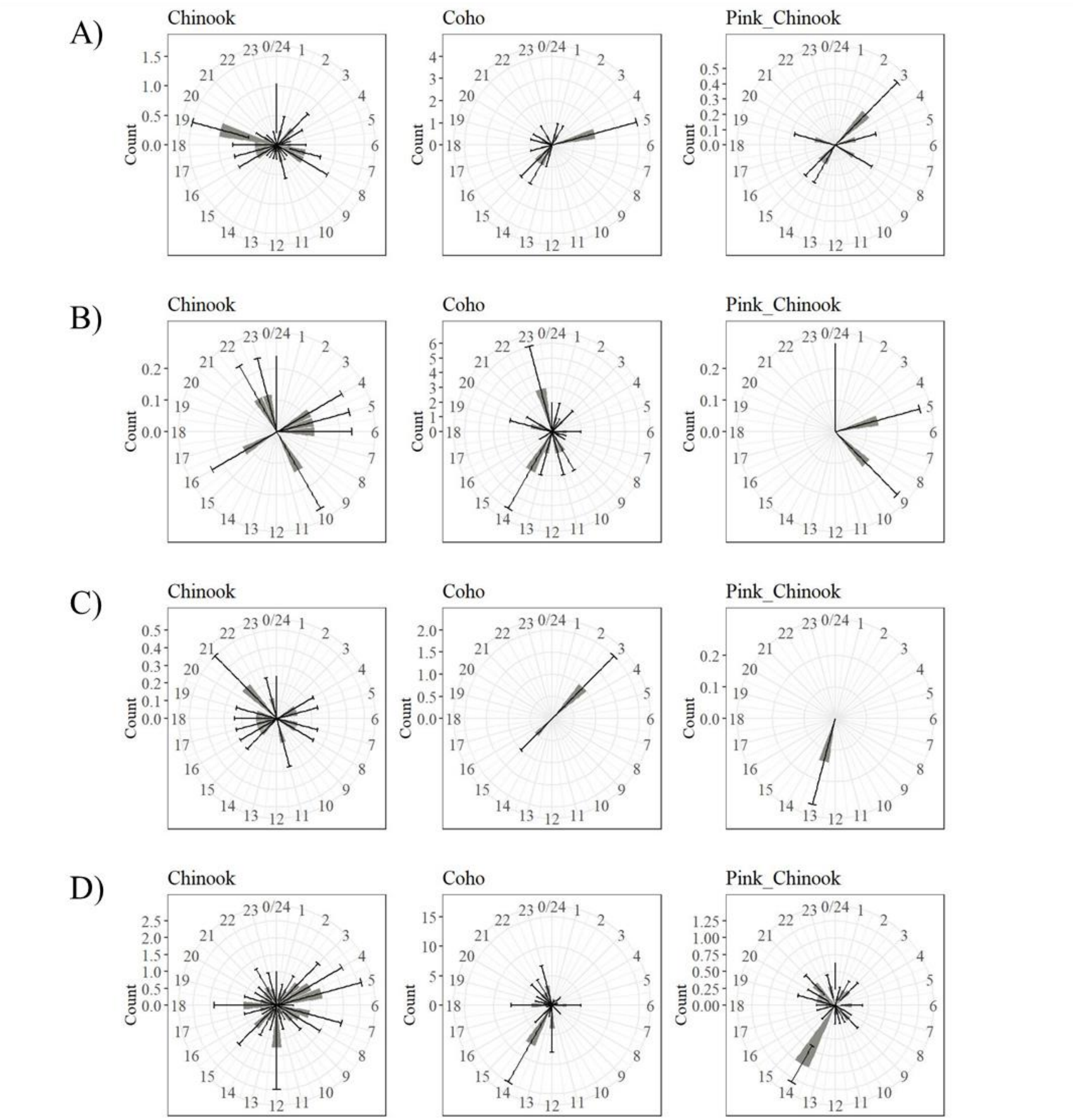

Figure B4.5. Circular bar graphs of mean and standard error of counts of sound production by hour of the day in each grouping of salmon, Chinook only ($n_1 = 8$), Coho salmon ($n_1 = 2$), Pink and Chinook mixed ($n_1 = 7$). A) Gill Bubble like sounds. B) Gurgle sounds. C) Moan sounds. D) Pulse sounds.

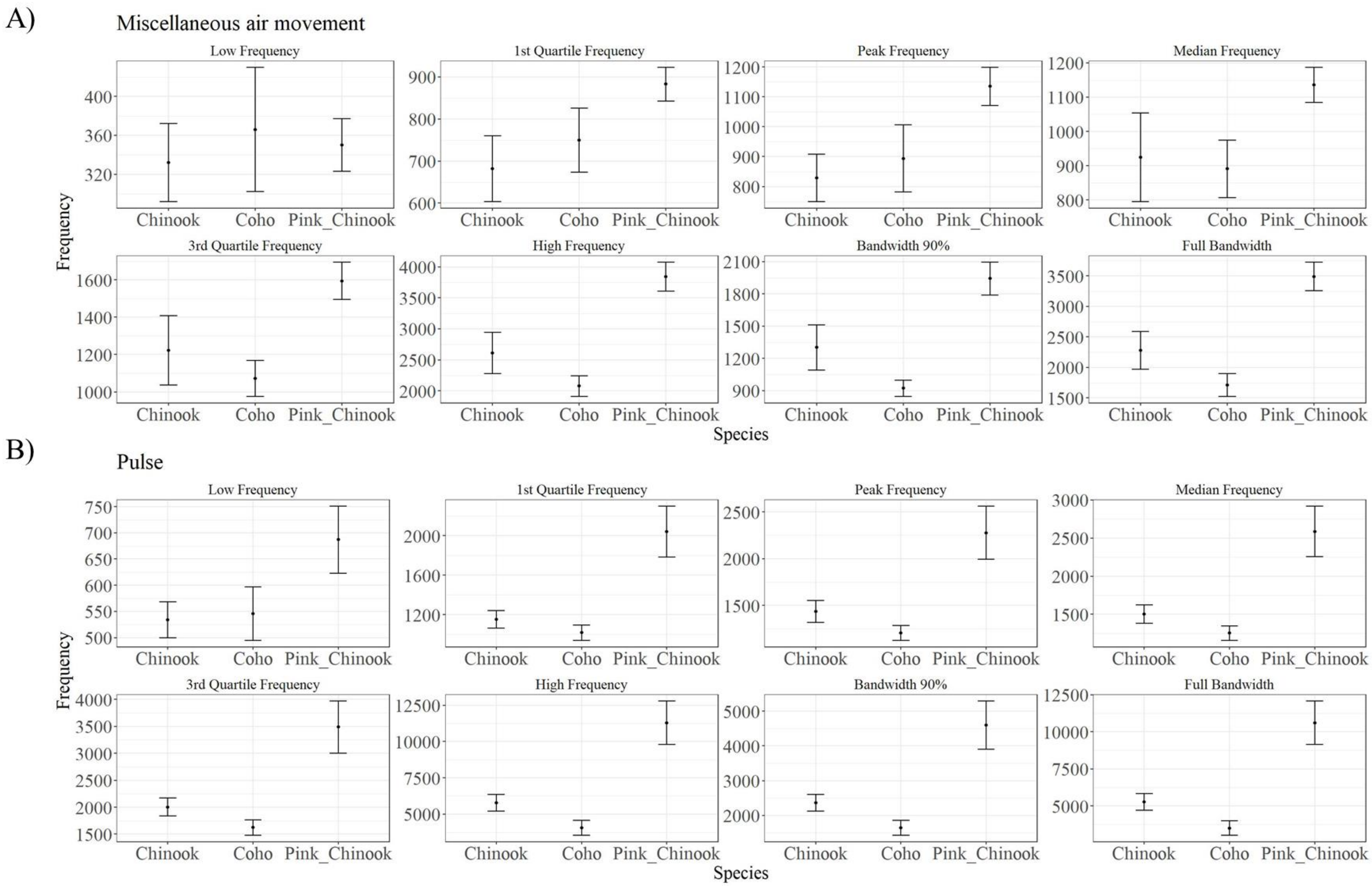

Figure B4.6: Mean ± SE for all frequency acoustic measurements for each species grouping. A) Miscellaneous air movement sounds. B) Pulse sounds

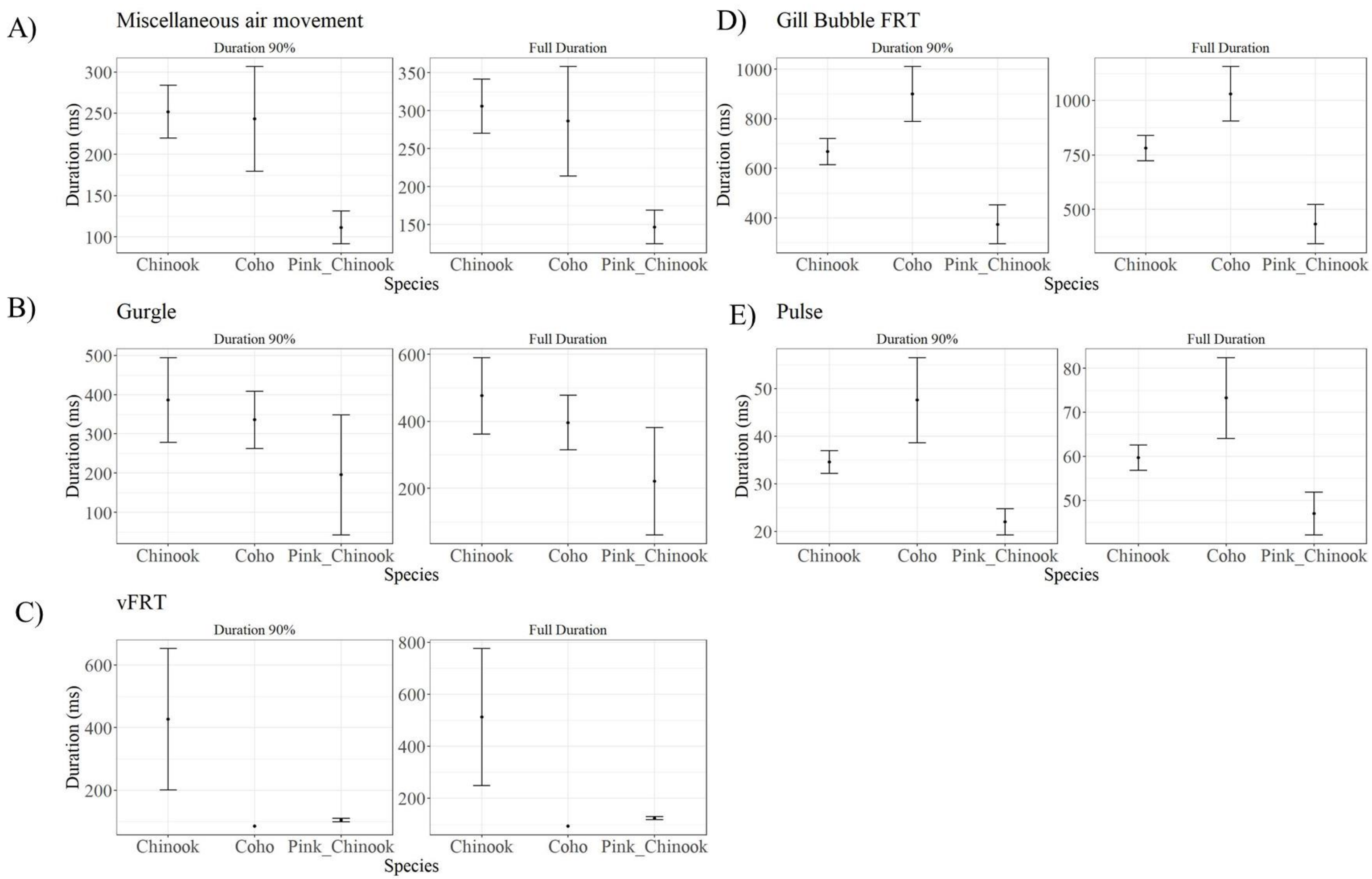

Figure B4.7: Mean ± SE for all duration acoustic measurements for each species grouping. A) Miscellaneous air movement sounds. B) Gurgle sounds C) vFRT sounds. D) Gill Bubble FRT sounds. E) Pulse sounds.

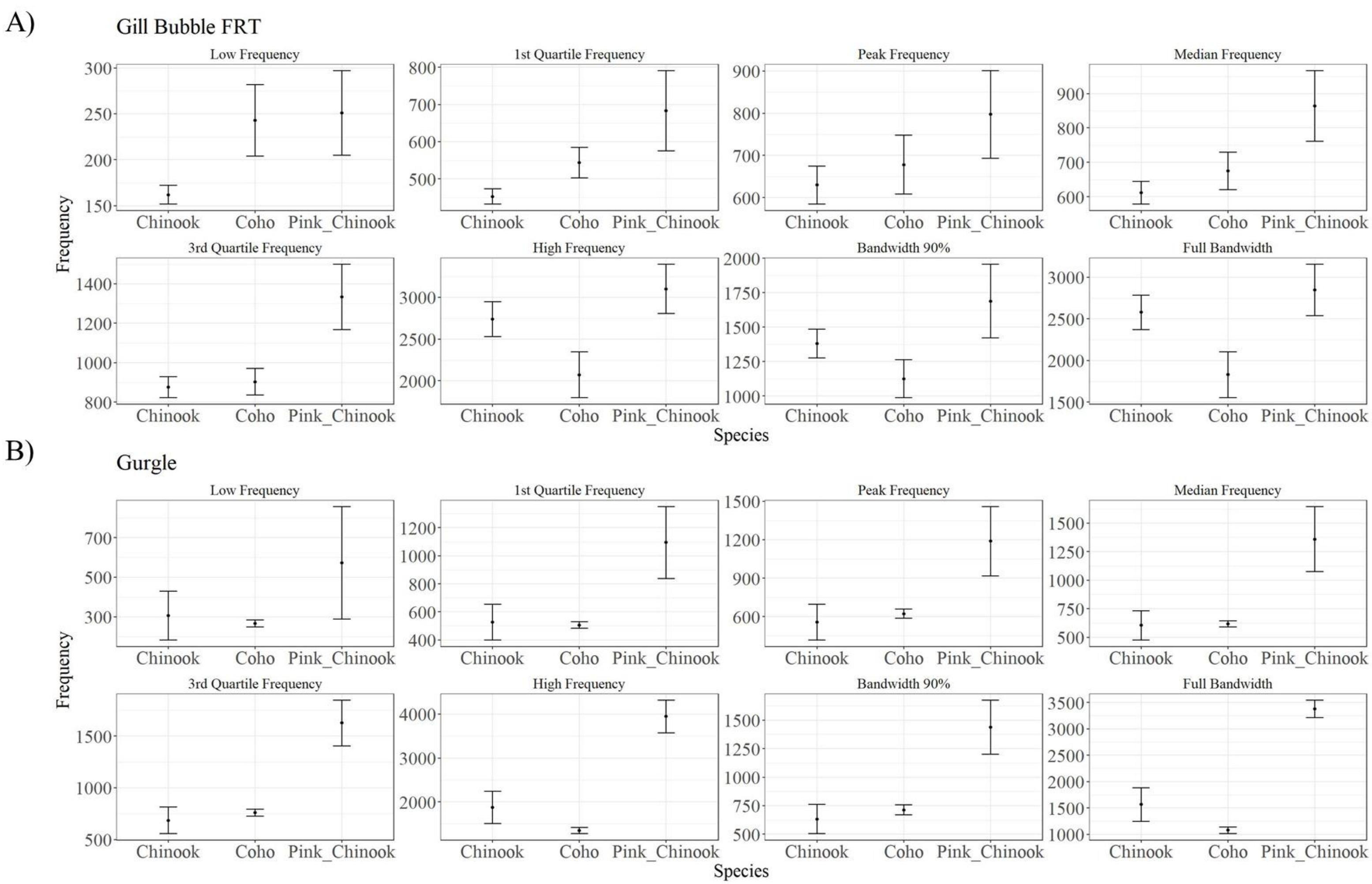

Figure B4.8: Mean ± SE for all frequency acoustic measurements for each species grouping. A) Gill bubble FRT sounds. B) Gurgle sounds.

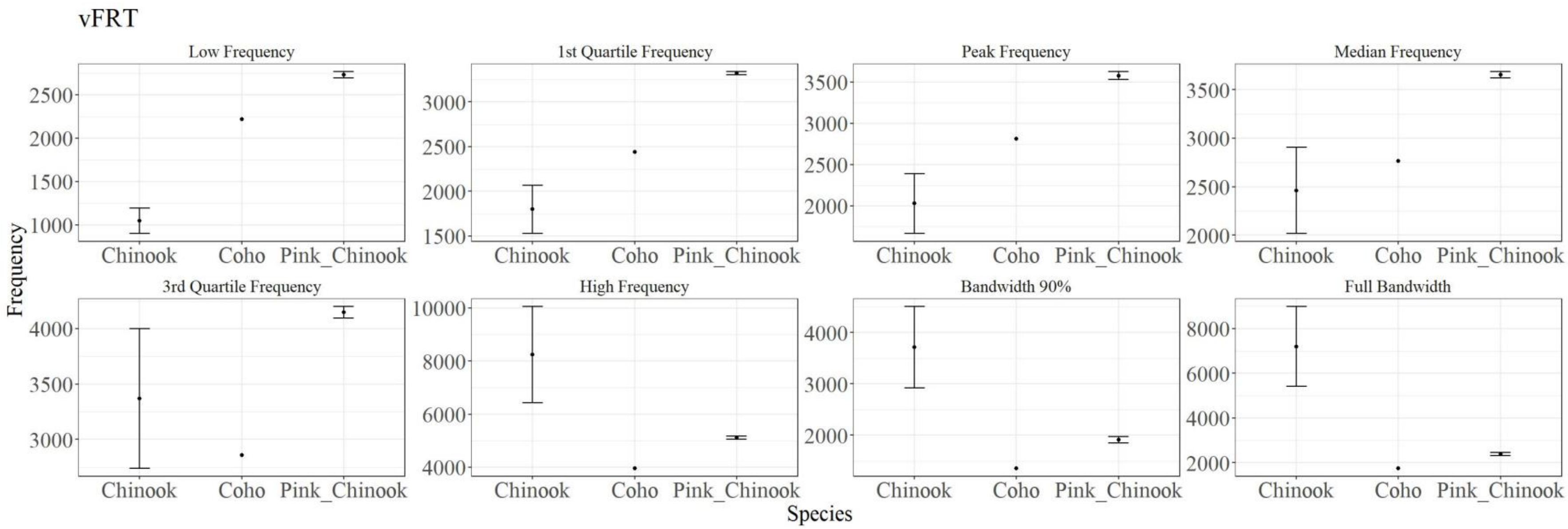

Figure B4.9: Mean ± SE for all frequency acoustic measurements of vFRT sound for each species grouping.

Appendix C: Supplemental Data for Chapter 5

Pilot study: Gastric tag retention

From September 25, 2018, to October 3, 2018, Chinook salmon (n = 48) were gastrically tagged using a plastic plunger at river km 2 in the Cowichan River. Each salmon was gastric tagged with a Teflon dummy tag (the same size and weight as the V13AP-1x acoustic tags) equipped with an FDX-PIT tag, and each salmon was also tagged with an HDX-PIT tag injected intraperitoneally. Tagged salmon had a mean fork length of 616.1 mm ± 129.5 mm. The sex ratio of tagged salmon was 21 females, 6 males and 21 of unknown sex, with 36 individuals of wild origin.

Tagged salmon were detected at both PIT tag arrays in the Cowichan River, the fence array at river km seven and the Skutz Falls array at river km 33. At the fence array, 41 individuals were detected 620 times. Salmon spent a mean time of 3.26 ± 7.14 days at the array and all individuals retained their gastric tag during that time; however, two individuals lost their intraperitoneal PIT tag. Fewer individuals made it to the Skutz Falls array, with 16 individuals being detected 32 times. Time spent also decreased at Skutz Falls, salmon only spent a mean time of 0.51 ± 1.70 minutes. All detected salmon had retained their gastric tag, but one more individual had lost its intraperitoneal tag. Tagged salmon took a mean time of 3.87 ± 1.14 days to travel the 26 km from the fence array to Skutz Falls. The last detection was on November 7, 2018.

Behaviour and Movement patterns outside Cowichan Bay

Movement patterns outside Cowichan Bay were also assessed for fish that were last detected leaving Cowichan Bay (last detected on receivers at the opening to Cowichan Bay) and never returned or were detected on nearby arrays. For all outside detections, mean ± SE of depth and activity values were tabulated for each year.

Each year some of the tagged salmon left the area and never returned, their final fates are unknown. Detection data from 2019 indicated that four acoustically tagged Chinook salmon left the bay and never returned (Tag IDs: 7942, 7948, 7952, 8216). Salmon 7942 (GSI: Cowichan River: probability 90%) was never detected again, but salmon 7948 (GSI: Cowichan River: probability 99%) was detected approximately 35 km away from the mouth of the Cowichan River on the Sidney Channel array (VEMCO; Bedford, NS; VR2W: Kintama/University of British Columbia Array) and never detected again. Salmon 8216 (GSI: Nitnat River: probability 97%) was detected on the Ocean Tracking Network Juan de Fuca line (VEMCO; Bedford, NS), while salmon 7952 (GSI: Handford Reach: probability 53%) was detected exiting the Strait of Georgia through Johnstone Strait, by the Johnstone Strait acoustic array (VEMCO; Bedford, NS; VR2TX: University of British Columbia Array), and was then detected entering Queen Charlotte Sound at the northern tip of Vancouver Island (VEMCO; Bedford, NS; VR2TX: Ocean Tracking Network). Nineteen days later Salmon 7952 was detected entering the Columbia River estuary, Washington, USA (VEMCO; Bedford, NS; VR2AR: WA/OR NOAA Array). Furthermore, Chinook 7952 was tracked up the Columbia River through PIT antennae at the Bonneville Dam (October 22, 2019), and McNary Dam (October 27, 2019), which is 236 km upstream. Additionally, one salmon (ID 7950, GIS: Nanaimo River (probability = 82%)) was also detected on the Sidney Channel array, returned to the Cowichan River, and was detected at the acoustic receiver installed at the mainstem array on October 3, 2019.

Similarly, four tagged (IDs 10576, 10578, 10612, 10616) salmon left Cowichan Bay in 2020 and were detected on acoustic receivers deployed by DFO (VEMCO; Beford, NS: VR2W). Salmon 10612 (GSI: Cowichan River: probability 99%) was detected closest to the mouth of the Cowichan River, 30 km away near North Pender Island (Swanson Channel), while salmon 10578

(GSI: Nitnat River: probability 99%) and 10616 (GSI: Chilliwack River: probability 96%) were detected along the west coast of Vancouver Island near Sooke and Port Renfrew, respectively. The last tagged salmon that left in 2020 (ID 10576, GSI: Soos Creek: probability 99%)) left shortly after tagging (<1 hour) and was never detected again.

In 2022, a higher proportion of tagged salmon left Cowichan Bay and never returned (7/19). Three tagged salmon (IDs 242, GSI: Cowichan River: probability 100%; 3976, GSI: Soos Creek: probability 99%; 12174, GSI: Unknown) were only detected on the Cowichan Bay array over multiple days before leaving and never being detected again. However, three tagged salmon were detected on DFO receivers in Boundary Pass (IDs 3966, GSI: Skagit River: probability 99%; 3970, GSI: Cowichan River: probability 99%), and Swanson Channel (ID 3972, GSI: Cowichan River: Probability 100%). Salmon 3966 was also detected on a DFO receiver in the Southern Strait of Georgia on October 25, 2022, then was detected in Swanson Channel on October 28th. An additional tagged salmon (ID 398; GSI: Green River: probability 66%) was detected on the west coast of Vancouver Island by the Jordan River on a DFO receiver and an Ocean Networks Canada receiver, before being detected on the Ocean Tracking Network Juan de Fuca line in early October 2022. Salmon 3980 was detected again in May 2023 on a DFO receiver in Haro Strait.

When tagged salmon were detected outside the Cowichan Bay array, differences in depth and activity values were detected among tagging years. Tagged salmon traveled at a mean ± SE depth of 32.2 ± 3.8 m in 2019, 51.4 ± 2.0 m in 2020, and 44.2 ± 2.0 m in 2022. All three tagging years had significantly different mean depths when traveling outside Cowichan Bay ($p < 0.001$). Mean activity values were the same in 2019 and 2020 (0.9 ± 0.03 m/s^2), however, in 2022 tagged salmon had significantly slower activity of 0.76 ± 0.03 m/s^2 ($p < 0.001$).

Time in the Estuary and River

Once in the Cowichan River, tagged Chinook salmon spent varying amounts of time at different river receivers. In the estuary, salmon spent the most time at the south arm estuary receiver in 2019 (mean ± se; 5.6 ± 4.9 hours), but only spent minimal time there in 2020 (0.6 ± 0.2 hours) and 2022 (0.2 ± 0.1 hours). Time spent at the north arm of the estuary was consistent among years (2019: 0.7 ± 0.2 hours; 2020: 1.7 ± 1.6 hours; 2022: 0.8 ± 0.7 hours), but longest in 2020. Salmon spent a wider range of time at the lower river receivers among years, with salmon spending the most time at the south arm receiver in 2019 (5.9 ± 2.6 hours) compared to 2020 (4.9 ± 1.9 hours) and 2022 (1.7 ± 0.6 hours), however, the pattern was reversed in the north arm, with salmon spending the most time there in 2022 (54.0 ± 30.8 hours) compared to 2020 (5.7 ± 4.3 hours) and 2019 (0.4 ± 0.3 hours). Yearly differences between which arm of the river salmon migrated up was also detected. In 2019 the majority of salmon (10/12) that moved upriver used the south arm, compared to 2020 where most (7/11) used the north arm and in 2022 where the same number of salmon moved up each arm (4). Finally, once salmon were in the mainstem of the river they moved quickly up to the fence in 2019 (Major Jimmy: 0.5 ± 0.3 hours; Fence: 3.5 ± 2.1 hours) and 2020 (Major Jimmy: 0.1 ± 0.02 hours; Fence: 0.8 ± 0.5 hours), but spent a considerable amount of time in 2022 at a deep pool in the river (Major Jimmy; 53.4 ± 50.0 hours), with one individual spending 18 days there, but moving faster past the fence (5.6 ± 4.1 hours).

Appendix D: Supplemental Table for Chapter 5

Table 5.1. Tagging data for Chinook salmon

Year	Date	Acoustic ID	Fork Length (mm)	Circumference (mm)	Stock ID	Percent	Origin	Sex	Maturity	Survived
2019	8/23/2019	7940	812	482	Nanaimo River (fall)	0.938	W	NA	NA	0
2019	8/24/2019	7942	840	490	Cowichan River	0.892	W	F	2	0
2019	8/24/2019	7944	750	420	Cowichan River	0.882	W	M	1	0
2019	8/25/2019	7946	710	420	Cowichan River	0.999	W	F	1	1
2019	8/26/2019	7948	850	510	Cowichan River	0.999	W	NA	1	0
2019	8/27/2019	7950	705	430	Nanaimo River (fall)	0.821	W	F	0	1
2019	8/28/2019	7952	840	465	Handford Reach	0.536	W	NA	0	0
2019	9/5/2019	7954	630	400	Cowichan River	0.999	W	NA	0	0
2019	9/5/2019	7956	835	464	Cowichan River	0.999	W	NA	1	1
2019	9/8/2019	8210	690	440	Cowichan River	0.999	W	M	1	1
2019	9/8/2019	8212	760	445	Cowichan River	0.999	W	F	1	0
2019	9/8/2019	8214	765	460	Cowichan River	0.999	W	M	2	1
2019	9/8/2019	8216	800	475	Nitnat	0.971	W	M	2	0
2019	9/11/2019	7958	630	350	Cowichan River	0.731	H	F	0	0
2019	9/11/2019	8218	830	485	Cowichan River	0.999	W	M	1	0
2019	9/19/2019	4634	815	470	Cowichan River	0.999	H	F	0	1
2019	9/19/2019	4636	755	450	Cowichan River	0.999	W	NA	1	0
2020	8/30/2020	10576	690	390	Soos Creek	0.999	H	M	2	0
2020	9/1/2020	10578	835	475	Nitnat	0.999	W	F	1	0
2020	9/2/2020	10580	755	430	Cowichan River	0.999	W	M	2	1
2020	9/3/2020	10582	850	470	Cowichan River	0.999	W	F	2	1
2020	9/4/2020	10584	840	460	Cowichan River	0.999	W	F	1	0
2020	9/5/2020	10606	790	415	Cowichan River	0.999	W	F	1	1
2020	9/5/2020	10608	760	450	Cowichan River	0.999	W	F	1	0
2020	9/8/2020	10610	805	480	Cowichan River	0.999	W	F	1	1
2020	9/9/2020	10612	910	540	Cowichan River	0.999	W	M	2	0
2020	9/12/2020	10614	750	460	Cowichan River	0.97	W	F	1	1
2020	9/14/2020	10616	725	412	Chilliwack River (fall)	0.96	W	M	1	0

2020	9/16/2020	10618	695	415	Cowichan River	1	H	F	1	1
2020	9/18/2020	10620	740	457	Cowichan River	0.999	W	F	0	0
2020	9/19/2020	10622	770	480	Cowichan River	0.999	W	M	2	1
2020	9/21/2020	10624	710	428	Cowichan River	0.89	W	F	1	1
2020	9/22/2020	12180	480	280	Cowichan River	1	H	M	0	1
2020	9/22/2020	12182	520	300	Cowichan River	0.999	W	M	0	1
2020	9/27/2020	12178	530	316	Cowichan River	0.999	W	M	1	1
2020	9/30/2020	12176	464	269	Soos Creek	0.999	H	M	0	0
2022	8/31/2022	234	888	505	Cowichan River	0.974	W	NA	2	1
2022	9/6/2022	240	770	440	Little Qualicum River	0.742	W	NA	1	0
2022	9/6/2022	12174	658	355	Unknown	NA	H	NA	1	0
2022	9/9/2022	242	590	340	Cowichan River	1	W	NA	2	0
2022	9/12/2022	236	740	400	Cowichan River	0.999	W	NA	2	1
2022	9/12/2022	3958	540	320	Cowichan River	1	W	NA	2	0
2022	9/15/2022	238	577	315	Cowichan River	0.999	W	NA	1	1
2022	9/15/2022	3956	680	380	Cowichan River	0.991	W	NA	2	1
2022	9/15/2022	3970	590	320	Cowichan River	0.999	W	NA	1	0
2022	9/15/2022	3972	705	410	Cowichan River	1	W	NA	2	0
2022	9/19/2022	3960	865	470	Cowichan River	0.991	W	NA	2	1
2022	9/19/2022	3964	675	375	Cowichan River	0.999	W	NA	2	1
2022	9/21/2022	3966	620	334	Skagit River	0.9999	H	NA	2	0
2022	9/21/2022	3974	709	408	Cowichan River	0.999	W	NA	3	1
2022	9/21/2022	3978	801	485	Cowichan River	0.997	W	NA	2	0
2022	9/21/2022	3980	645	349	Green River	0.665	H	NA	2	0
2022	9/22/2022	3968	738	395	Nanaimo River (fall)	0.693	W	NA	2	1
2022	9/26/2022	3962	770	420	Cowichan River	0.998	W	NA	1	0
2022	9/26/2022	3976	792	425	Soos Creek	0.974	H	NA	3	0
2022	9/26/2022	3982	720	385	Cowichan River	0.998	W	NA	2	0

Appendix E: Supplemental Figures for Chapter 5

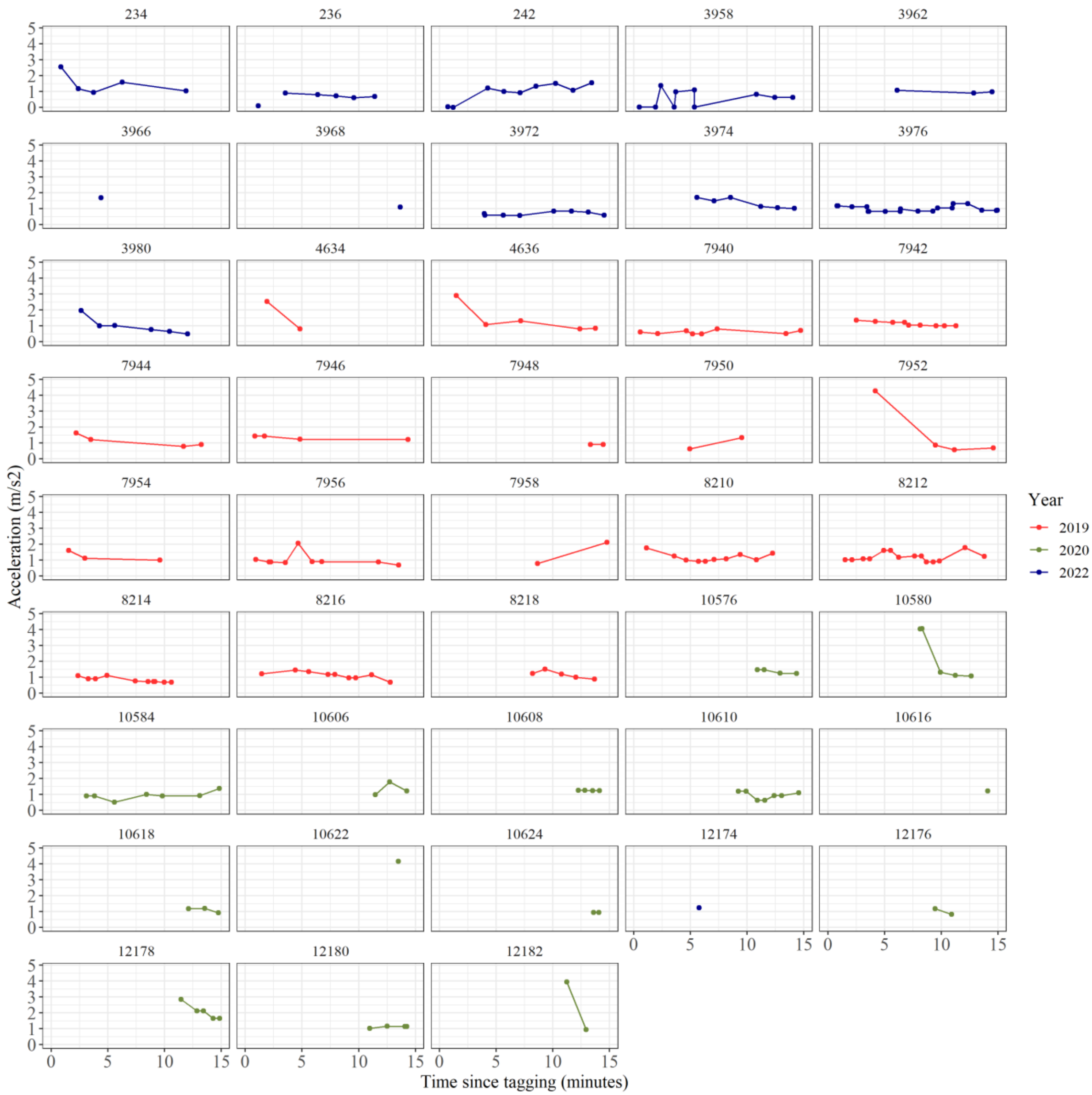

Figure E5.1. Activity data of acoustically tagged Cowichan Chinook. A mobile VR100 and a VR2 subsurface array collected data during post tagging releases for up to 15 minutes.

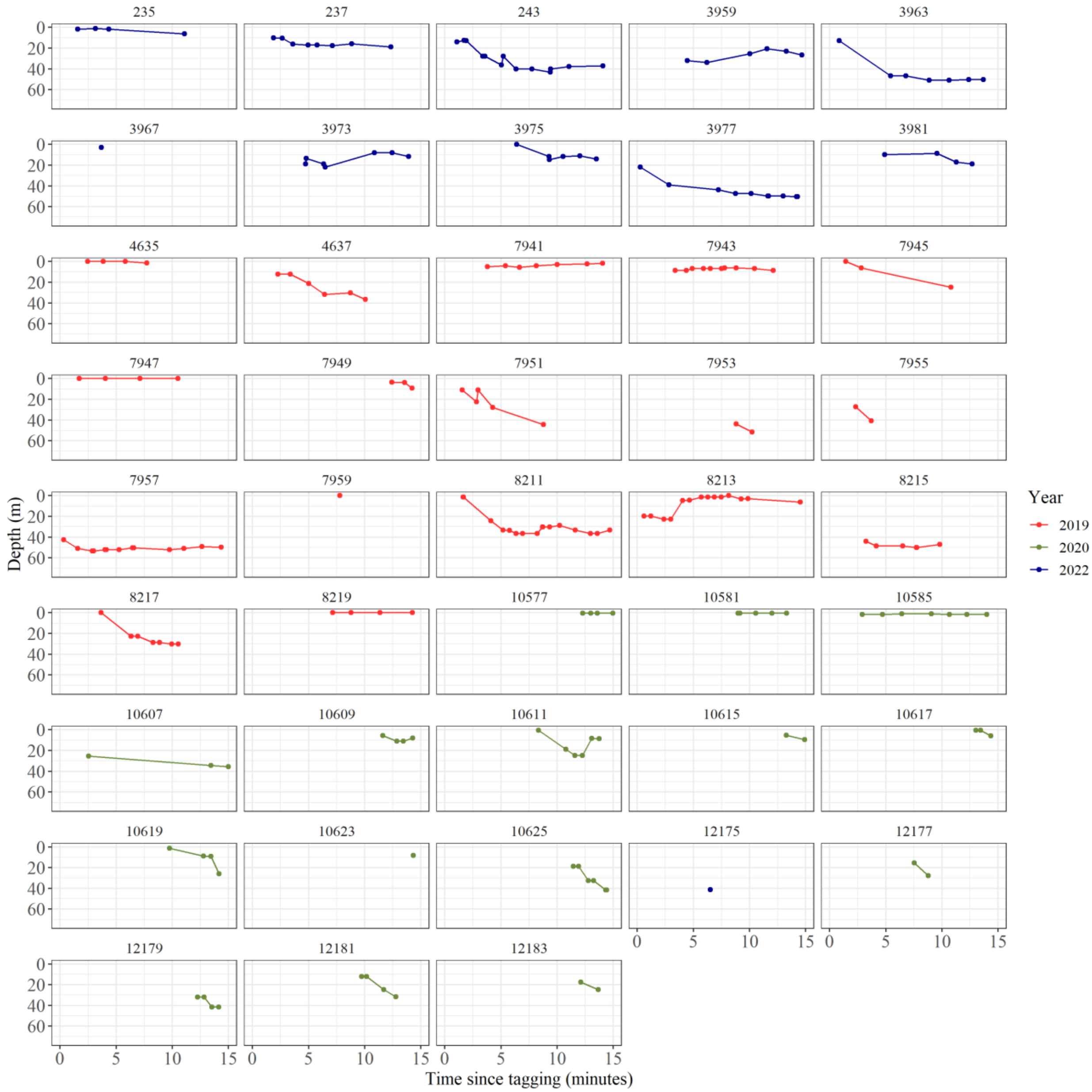

Figure E5.2. Depth data of acoustically tagged Cowichan Chinook. A mobile VR100 and a VR2 subsurface array collected data during post tagging releases for up to 15 minutes.