Evidence of Temporary Threshold Shifts in the Sub-Population, J-Pod, of Southern Resident Killer Whales Induced by Large Vessels in Haro Strait.

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It is becoming more and more apparent that humans are influencing multiple aspects of endangered southern resident killer whale (SRKW) life history (Ylitalo et al., 2001; Holt, 2008, etc.) Acoustic disturbances are among one of the most obvious and harmful immediate effects with short and long-term repercussions. Anthropogenic noise pollution can compromise SRKWs in a variety of ways. The nature of SRKW's acoustic repertoire consisting of pulsed and tonal calls called whistles (Thomsen et al., 2001), combined with the overwhelming presence of many whale watch vessels, as well as other vessels, causes masking to occur; this event impedes important aspects of daily life such as foraging and communicating with con-specifics (Foote et al., 2004). As a result, the SRKWs are forced to communicate louder (exhibiting a Lombard Effect), longer (Foote et al., 2004), adopt less predictable paths of movement (Williams et al., 2002; Holt, 2008; Goold and Fish, 1998) and endure compromised immune system levels due to stress (Simmons and Dolman, 2000.)

Elevated levels of noise pollution have further been proven to cause temporary hearing loss or temporary threshold shifts (TTS). This involves a temporary loss of baseline hearing after a noisy event takes place, fatiguing the inner ear hair cells and resulting in the loss of ability to hear lower amplitude sounds. The magnitude of the threshold shift is dependent upon amplitude, duration, temporal pattern, frequency, and energy content of sound (Holt, 2008).

TTS has not been monitored in killer whales as inducing such events is not feasible for a wild population, especially one that is endangered, though studies have been conducted on captive animals. The most closely related species to killer whales which have been studied in captivity are the beluga whale and bottle nose dolphin. These two species provide the best data available to establish threshold levels of permanent and temporary hearing loss from sound exposure in killer whales (Holt, 2008). Exposure to continuous noise or tones (non-impulsive sounds) in bottlenose dolphins and beluga whales indicate that, quite often, significant threshold shifts are seen at sound exposure levels (SEL) greater than or equal to 195 dB re 1 $\mu Pa^2 s$ (Holt, 2008). According to Fineran et al. (2005), this will occur despite differences in exposure duration, sound pressure level (SPL), experimental approaches, and subjects. More recently, Fineran et al. (2007) established that the largest hearing threshold shifts seen in bottlenose dolphins and beluga whales established so far are observable at 10, 50, 60 and 70 kHz. Au et al. (1999) have also done considerable work investigating TTS in captive bottlenose dolphins (*Tursiops truncatus)*. In a particular study by Natchtigall et al (2003), a bottlenose dolphin was exposed to octave band noise between five and 10 kHz for 30-50 minutes. It was discovered that the auditory threshold post-exposure had shifted 96 dB above the normal central frequency. Erbe (2002) used Au et al.'s 1999 data and scaled down the TTS using human and other terrestrial animals to become comparable to the killer whale bandwidth range. By doing this, she found that an exposure to 68-74 dB octave band level above audibility is assumed to result in a five dB TTS in delphinids after 30-50 minutes of exposure. Erbe (2002) further pursued this topic by using preliminary data from a study conducted by Nachtigall et al. (2003) in which a TTS of 12-18 dB at 7.5 kHz was induced in a bottlenose dolphin after exposure to an octave band of noise at 179 dB re 1μPa for approximately 50 minutes. Schlundt *et al.* (2000) further confirmed that cetaceans experience TTS in their own study of bottlenose dolphins examining masked temporary threshold shift (MTTS-in background noise) after exposure to pure tones. With the fatiguing stimulus at 3 kHz, they measured a MTTS of 7 dB at 3 kHz, 16 dB at 4.5 kHz (half an octave above the noise frequency), and 17 dB at 6 kHz (one octave above the noise frequency).

The above investigators of TTS in marine mammals, especially cetaceans, have proven that such events can take place at realistic dB and frequency levels in captive bottlenose dolphins and beluga whales. Holt (2008) points out, however, that hearing sensitivity is slightly better in killer whales than bottlenose dolphins between four and 20 kHz. Keeping this in mind, Veirs & Veirs (2007) found that over an 18 month period the average background broadband noise emitted from large commercial ships in Haro Strait, Puget Sound was 144 dB re 1µPa over the 100 Hz – 15 kHz bandwidth. If large, slow-moving container ships are emitting noise levels at 144 dB near SRKWs with a sensitive hearing range just under 144 dB for durations which last approximately an hour (Veirs, personal communication), it is quite conceivable that they are experiencing TTS. Further, Holt (2008) states that if SRKWs experience these levels of sound exposure for eight hours a day, five days a week for five years, they would acquire permanent hearing loss.

In this study I will determine if Southern Resident killer whales are experiencing TTSs as a result of large vessel traffic (container ships, cargo ships, oil rigs). I hypothesize that the large vessel traffic remains loud enough for long enough in the SRKW's hearing range for TTS to occur based on the data discussed above. If this can be established, it could have management implications such as the potential to bind federally operated vessels to state regulations; the Be Whale Wise regulations require state-operated commercial vessels as well as private boaters to reduce their travel speed to seven knots when within 400 meters of any killer whales and put their vessels in neutral at 100 meters away. Such vessels are also required to approach the whales no closer than 100 meters. These guidelines do not prevent acoustic impacts, but similar regulations geared towards protecting the whales acoustically from large vessel traffic might need to be established if TTS events are occurring. Establishing if SRKWs are experiencing TTS would also shed further insight into the kind of acoustic habitat they live in as well as how humans may be compromising it. Finally, and most importantly, it is necessary to establish if SRKWs are experiencing TTS as it could have more serious repercussions in the future, such as permanent threshold shifts (PTS.) PTS is a worst case scenario and is well worth investigating in an effort to establish if it could happen in the future. Providing data which can help establish policy regulations and provide a more clear understanding of the acoustic environment SRKWs are living in by using the best and most current information available is essential to ensuring their existence.

Methods:

The study of J-Pod's amplitude of sound in response to large vessel traffic will be conducted predominantly in Haro strait, Washington state, U.S.A, but will extend to adjacent waters if the whales travel there as well, potentially including Canadian waters. Data will be collected from the research platform the Gato verde, a 42-foot sailing catamaran. This vessel, when not operating under wind power, operates by using two electric propulsion motors run from battery banks charged with a bio-diesel generator. This is an important aspect of the vessel as it does not add sound to the acoustic environment being studied and thus allows optimal conditions for a research platform to work in the field without interference.

To establish whether or not there is evidence that J-Pod is experiencing temporary threshold shifts (TTS) induced by large vessels, sound amplitude from the source, or source level (SL) before and after exposure will be compared. A program called Nobeltech Admiral Navigation, a marine navigation software, will aid in detecting large vessels before they enter the vicinity where they could likely cause TTSs. This will allow ample time to ensure that the hydrophone arrays for recording vocalizations are in the water for the appropriate amount of time. This software will also assure that other logistical aspects necessary to this study are in place before the vessel arrives. Further, the software will log the large vessel's distance from the Gato Verde throughout the extent of its presence near the whales. This can help verify localizing outputs from the Ishmael software.

When J-Pod is encountered, a hydrophone array consisting of three Lab-core hydrophones with peak sensitivity of 5,000 Hz (down 30 dB at 200 and 10,500 Hz) will be deployed. Underwater sound will be recorded using two solid state recorders with a proprietary link for sample accuracy. The solid state recorders are Sound Devices 702 with a flat frequency response from 10 Hz to 40 kHz (+0.1,-0.5 dB). Recording J-pod vocalizations pre- and post-exposure will be accomplished by using a towed array hydrophone deployed off the port stern pulpit of the Gato

Verde. The array is depicted in figure 1 below:

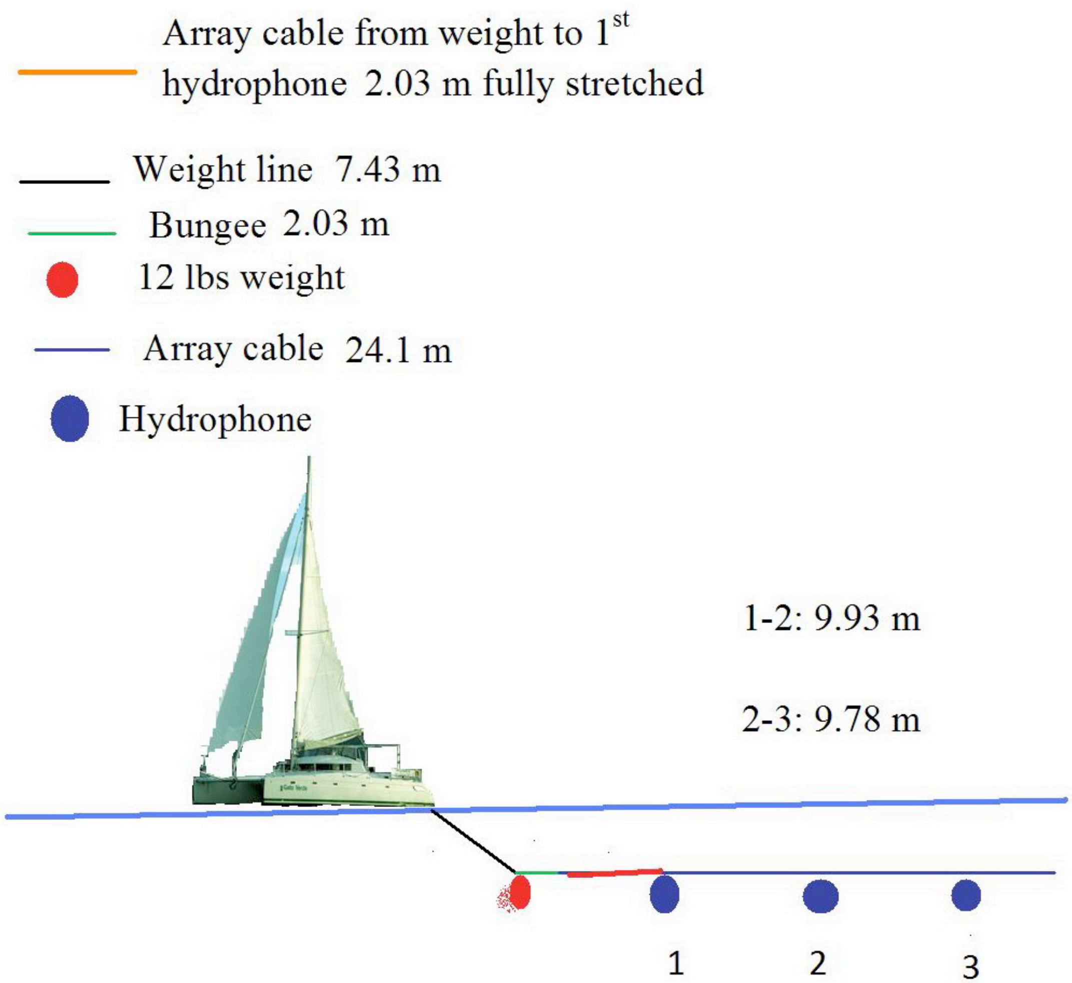

Figure 1: diagram of the Gato Verde with the towed array and the positions of its various elements.

In addition, off the starboard stern pulpit a high frequency hydrophone will be towed in order to eliminate ambiguous positions while localizing calls.

For the purpose of this study, only S1 calls will be examined in order to maintain consistency and directly comparable data between pre- and post-exposure periods. This call was chosen as it is J-pod's signature call and is used, on average, 50% of the time despite their large call repertoire (Ford 1989, Ford 1991). For this study, one data set will be considered a recording of pre-exposure vocalizations to large vessel noise as well as a recording of post-exposure vocalizations, both containing, ideally, at least 10 S1 calls for analysis. The pre- and post-exposure periods will ideally last 15 minutes, however each particular situation will have different time constraints and thus the pre- and post-exposure periods may have to be adjusted accordingly. Adjustments may also be necessary if an insufficient amount of S1 calls are made in the time allotted for pre- and post-exposure periods. Pre-exposure periods for collecting S1 calls are much more flexible and acceptable to collect calls outside of this time. However, post-exposure should remain relatively short and consistent as recovery period is uncertain; waiting too long could result in measuring calls which have already returned to baseline level. The exposure period will begin when the large vessel is heard on the speakers connected to the hydrophones. Though this may seem ambiguous, detecting a consistent start period is difficult to establish without first experiencing it. The end of exposure period will be determined by calculating and graphing the SEL of the ship over the period which it is present at intervals of eight minutes. At this interval, a model can be generated in an excel spreadsheet which can detect the pattern the SEL will take on and thus cessation of exposure period can accurately occur at 90%. Once the S1 calls are recorded, SL will be measured in order to compare amplitudes of sound before and after vessel exposure. To begin this process, the S1 calls will be selected from each recording by examining spectrograms displaying frequency on the y-axis and time on the x-axis in a program called Raven Lite. Each call will first be opened with the localization program Ishmael (David Mellinger), which detects differences in sound between the hydrophones of the array to figure out the distance between detector and sound emission. Entered into this software will be the distances of each hydrophone from each other and their placements within the array off the Gato Verde's stern, the sound files themselves, and a speed of sound measurement of 1480 m/s. Ishmael can then combine all the information to calculate the x and y values making up the opposite and adjacent sides of a triangle. This makes up the boat's position, the hydrophone array, and the whale. With these two pieces of information, Pythagorean's theorem ($A^2+B^2=C^2$) can be used to calculate the hypotenuse, or the distance from the vocalizing whale to the Gato verde. This information will be used to calculate the loss of sound, or spreading loss which can be expected for each particular distance. The formula $20*\log_{10}(R/R_0)$, where R is the range or distance calculated in Ishmael and R_0 is the reference range of 1 meter, all assuming a spherical spreading loss model. The result of this equation will then be added to the RL, which can be calculated in the program OVAL (V. Veirs, 2008), to calculate SL.

The sensitivities used for calculating RMS and RL in OVAL are listed in the table below:

Sensitive RMS for each channel:		
		Sensitive RMS
Channel	1	143
	2	149
	3	145
	4	147

Figure 2: table depicting sensitive RMS for each of the recording channels on the sound devices

Calibrating the hydrophones to get individual sensitivities is necessary in order to record amplitude of sound in dB re 1µPa and therefore directly comparable to the work of other researchers working in this field.

The following table lists other pertinent settings on the sound devices which will be used during recordings. The low-cut filter will be used to block any noise levels listed below to keep drag of water against the hydrophones from being recorded, as well as any other ambient noise which could negatively affect the signal to noise ratio.

Gain settings for each channel					
			Bit depth	Sampling ra	Low-cut filt
		Gain settir	16	192	240 Hz
Channel	CRT-----1	28.4			24 dB/octa
	2	36.7			
	3	27.1			
	4	33.1			

The appropriate statistical test which will be used to assess the study's probability of correctly rejecting the null hypothesis will be a repeated measures ANOVA. This will test the quality of means. However, in the event that J-Pod members alter their behavior in the middle of a pre- or post-exposure recording session, the changes will be noted as this could change the amplitude at which they emit S1 calls. In order to compensate for changes in behavior, a covariate analyses will be necessary to ensure that this independent variable is not affecting the primary interest variables, or the relationship between the dependant variable (dB of S1 calls) and independent variable (before and after exposure periods).

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