Final Report

Assessment of Vocal Behavior of Sperm Whales in the Northwestern Atlantic Ocean

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Sperm whale (*Physeter macrocephalus*) photograph taken by UNCW under NOAA Permit Number 948-1692-00.

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Executive Summary

In this study, we conducted a detailed analysis of existing passive acoustic data using a combination of automated and semi-automated methods to characterize the vocal behavior of sperm whales (*Physeter macrocephalus*) in the middle to southern U.S. coastal Atlantic Ocean. Existing datasets from autonomous recorders deployed offshore of Jacksonville (JAX), Florida, Onslow Bay (OB), North Carolina, and Cape Hatteras (HAT), North Carolina, were analyzed to assess the presence, foraging behavior, and diel patterns of sperm whale vocal activity in these regions. The dataset used for this study consisted of acoustic data from ten autonomous recorders: nine Marine Autonomous Recording Units (MARU's) [six JAX (three fall and three winter), three OB] and one Autonomous Multi-channel Acoustic Recorder (AMAR; HAT).

For each autonomous recorder in the dataset, long-term spectral averages (LTSAs) were created using the MATLAB-based program Triton (Wiggins 2007). These LTSAs then were used to log (i.e., annotate) sperm whale acoustic encounters, defined as continuous periods of time containing sperm whale echolocation clicks, with no more than a 30-minute interval between echolocation clicks. Next, all .wav files containing sperm whale encounters were post-processed using an automated echolocation click detector in PAMGuard software (Gillespie et al. 2008). Each sperm whale encounter was then further processed using PAMGuard's Viewer Mode software to mark click train and foraging buzz events in the dataset. Echolocation clicks were then exported to the Real-time Odontocete Call Classification Algorithm (ROCCA), a module in PAMGuard, which was used to obtain click counts and echolocation click measurements including duration (microseconds), center and peak frequency (kilohertz), number of zero crossings, sweep rate (kilohertz/millisecond), and inter-click-interval (ICI; seconds). We performed a randomization test (10,000 replicates) of the Analysis of Variance (ANOVA) F-statistic to determine if there were significant differences in echolocation click measures among sites and examined pairwise median differences to compare measures between sites. The among-region comparisons showed that all regular click-measure parameters were significantly different. However, pairwise comparisons showed that peak frequency was significantly different only at OB compared to all other deployments. Sperm whale occurrence was plotted from the encounter logs by day and also by time of day for each recorder to provide an overview of vocal activity for each recording instrument. For each recorder, we calculated the number of click trains and the number of foraging buzzes, the number of days with click trains and the number of days with foraging buzzes, the number of click trains per day, the total duration of click trains and the total duration of foraging buzzes, the percentage of days with click trains and the percentage of days with foraging buzzes, the proportion of time vocally active and the proportion of vocalization time spent in prey capture attempts. G-tests were performed on contingency tables of the following variables to test the null hypothesis that each of the following variables is independent of region: 1) number of days with (and without) clicks: 2) number of days with (and without) buzzes: 3) number of vocalizations that are regular clicks (versus foraging buzzes); and 4) the number of seconds spent clicking (versus buzzing). The two JAX deployments (JAX-1 and JAX-2) were examined in more detail by executing the G-test on the same variables by only these two deployments.

Vocal behavior varied both within and among the deployments compared. Overall the highest number of encounters and highest total duration of encounters occurred during JAX-1 (n=90 encounters, approximately 113 hours [hr]), followed by JAX- 2 (n=46 encounters, approximately 68 hr), OB (n=39 encounters, approximately 50 hr), and HAT (n=28 encounters, approximately 23.5 hr). The proportion of days with click trains and foraging buzzes present was highest overall during the JAX deployments followed by HAT and OB. The number of click trains per day was highest for JAX-1 (n=22), followed by OB (n=18), JAX-2 (n=17), and HAT (n=5), respectively. The number of foraging buzzes per day was highest overall for JAX-2. The percentage of recording days with vocal activity varied by both recording site and geographic region/deployment. Click trains were present every day of the JAX-2-6 recorder deployment, 91 percent of days at OB site 1-2, and 83 percent of days at JAX-1-4. Foraging buzzes were present during the highest percentage of days overall during both JAX deployments (10 percent) followed by HAT (3 percent), and OB (1 percent).

The proportion of days with clicks was significantly different from that expected among all regions if it were independent of region, but there was no significant difference in the proportion of days with clicks between the two JAX deployments (i.e., fall versus winter). The proportion of days with foraging buzzes was not significantly different from that expected given independence among regions or between the two JAX deployments. The total number of clicks compared to the total number of foraging buzzes and the number of seconds clicking versus buzzing was found not to be independent of region or season (JAX).

Diel patterns in the occurrence of vocal events at each site were examined by dividing the recordings into 3-hour and 1-hour time bins as well as photoperiods (i.e., light versus dark) and obtaining click counts for each period. A Kruskal-Wallis test was then used to determine if there were significant differences in the number of clicks among: (1) 3-hour time bins within and among sites; (2) photoperiods within and among sites; and (3) hourly time bins within and among sites. We also performed multiple comparison Dunn's tests with Bonferroni corrections to determine how diel patterns varied between sites. Click counts were found to be significantly different among 3-hour time bins and 1-hour time bins and between photoperiods at every site except HAT, and were significantly different (p< 0.05) among all sites overall.

The interpretation and discussion of results from this analysis must be prefaced by acknowledging several caveats. First, there was limited spatial sampling at each of the three study regions. Second, there was temporal variation in sampling whereby JAX was sampled in fall and winter, OB was sampled in summer, and HAT was sampled in winter. Finally, the sample size of foraging buzzes was lower than expected. Despite these caveats, this study provides new information about the distribution, occurrence, and vocal behaviors of sperm whales in the coastal northwestern Atlantic. The results address gaps in current knowledge of sperm whale occurrence and behaviors, including the persistent presence, occurrence of foraging activity, and vocal behaviors of these deep-diving marine mammals in regions where they have been very rarely sighted using traditional visual methods. Additional sampling using both passive acoustic methods, such as towed-hydrophone-array surveys and tracking, coupled with electronic (e.g., satellite) tagging are required to provide more information about the occurrence and activities of sperm whales in the study regions.

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- Figure A-1. PAMGuard click detector display showing the bearing (y-axis) versus time (x-axis) display with detected clicks represented as filled shapes with the color indicating automatic classification of species or species groups. Using the 'semi-automated method,' selected clicks can be manually assigned by the user to a 'whale train,' which is then sent to ROCCA for measurement. In contrast, in the 'automated method' all clicks colored as the species of interest (e.g., beaked whale (orange) would be sent to ROCCA for measurement).
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Abbreviations and Acronyms

AMAR Autonomous Multi-channel Acoustic Recorder

ANOVA Analysis Of Variance

CV Coefficient of Variation

dB decibel

JAX Jacksonville

ESA Endangered Species Act

HAT Cape Hatteras

hr hour(s)

Hz Hertz

ICI inter-click interval

kHz kilohertz

km kilometer(s)

LTSA Long Term Spectral Average

m meter(s)

MARU Marine Autonomous Recording Unit

MMPA Marine Mammal Protection Act

μPa micro Pascal

μs microsecond(s)

ms millisecond(s)

NOAA National Oceanic and Atmospheric Administration

OB Onslow Bay

OBIS-SEAMAP Ocean Biogeographic Information System Spatial Ecological Analysis of

Megavertebrate Populations

ROCCA Real-time Odontocete Call Classification Algorithm

sec second(s)

USWTR Undersea Warfare Training Range

U.S. United States

.wav Waveform Audio File Format

1. Introduction

Sperm whales (*Physeter macrocephalus*) are large, deep-diving cetaceans with a cosmopolitan distribution. They inhabit deep waters in all major ocean basins, from the tropics to the polar regions (Rice 1998). Presently, this species is listed as endangered under the Endangered Species Act (ESA) and depleted under the Marine Mammal Protection Act (MMPA; NMFS 2010). In the western North Atlantic, sperm whales occur in waters over the continental shelf, slope, and abyssal plain (Smith et al. 1996; Davis et al. 2002; Hamazaki 2002; Waring et al. 2001, 2015). Studies of sperm whale habitat preference provide a link between the species' occurrence and prey availability in offshore waters. In particular, there is conjunction with topographical features associated with high primary and secondary productivity, such as the shelf break/slope and seamounts (Aissi et al. 2012, Mussi et al. 2014, Pirotta et al. 2011, Sagnol et al. 2014, Wong and Whitehead 2014). Research along the United States (U.S.) Atlantic coast also has linked the spatio-temporal distribution of sperm whales with productive waters along the shelf break at night (Hain et al. 1985, Hodge et al. 2013).

The distribution of sperm whales along some areas of the middle to southern U.S. Atlantic coast is still poorly documented despite decades of aerial and shipboard survey effort (Rickard 2015). Aerial surveys conducted during January 2009 through May 2012 over the Jacksonville FL area resulted in only one sighting of two sperm whales in offshore waters (approximately 265 meters depth) (DoN 2010, DoN 2013). Shipboard and aerial surveys conducted during June through August 2011 from central Florida to the lower Bay of Fundy resulted in a combined abundance estimate of 2,288 (Coefficient of Variation (CV) = 0.28) sperm whales in this region; 1,593 (CV = 0.36) estimated from aerial/shipboard surveys from central Virginia to the lower Bay of Fundy, and 695 (CV = 0.39) estimated from a concurrent shipboard survey from central Florida to central Virginia (Palka 2012). In addition, analysis of acoustic data collected with Marine Autonomous Recording Units (MARUs) by Norris et al. (2012) characterized the occurrence of sperm whales off the coast of Jacksonville based on vocalization events recorded during the fall/winter 2009 and winter 2010. The occurrence of sperm whale clicks also was documented using a 2008 MARU dataset from Onslow Bay, North Carolina (Hodge et al. 2013). In both of these studies, sperm whale clicks were detected most frequently by the mid-depth recorders between dusk and dawn. In Jacksonville, because sperm whales were not detected on offshore, deep water recorders, it was suggested that animals may be traversing through the study region along the shelf break to forage (Norris et al. 2012). Norris et al. (2012) also suggest that sperm whales in Jacksonville may have strong diel foraging and habitat preferences; however, the lack of click detections during the day does not necessarily mean that sperm whales are not present during this time, only that they are not acoustically active. Additional information is needed in order to better understand the observed patterns in sperm whale occurrence and to elucidate the behavioral ecology and habitat use of this deep-diving species within East Coast Navy ranges.

Sperm whales produce distinctive, broadband (100 Hertz [Hz] to 25 kilohertz [kHz]) echolocation clicks for approximately 80 percent of the time while they are actively diving (Miller et al. 2008). These 'click trains' are frequently characterized by evenly spaced pulses of decaying amplitude, although the pulse repetition rate can vary (Backus and Schevill 1966). Adult male sperm

whales have been observed to produce low-frequency, high-intensity clicks termed 'slow clicks' or 'clangs' that can reach source levels of 223 dB re: 1 μPa @ 1 m for adult males (Gordon 1987, Møhl et al. 2000). These distinctive clicks have not been observed from groups of females/juveniles. All sperm whales produce 'regular' clicks (also referred to as 'usual' clicks) that have a 0.5- to 1.0-second inter-click interval (ICI) and centroid frequency of 15 kHz (Madsen et al. 2002). Regular clicks are produced ubiquitously by sperm whales during dives and can be used to track and monitor their presence when they are not at the surface. During presumed foraging dives, sperm whales emit both "regular" clicks and "buzzes" (Whitehead 2003). Buzzes are a type of click train consisting of a click sequences with short inter-click intervals (0.2-0.5 seconds) that decrease during the course of the buzz (Jaquet et al. 2001, Watwood et al. 2006). Foraging buzzes consist of a long series of regular clicks, interspersed with short periods of rapid clicks called buzzes or creaks (Miller et al. 2004).

In this study, we conducted a detailed analysis of existing passive acoustic data using a combination of automated and semi-automated methods to provide detailed information about the vocal behavior of sperm whales in the coastal waters off the middle to southern U.S. Atlantic Ocean. Existing datasets from Jacksonville (JAX), Florida, Onslow Bay (OB), North Carolina (OB), and Cape Hatteras (HAT), North Carolina were analyzed to assess the presence, foraging behavior, and diel patterns of sperm whales in these regions. Foraging buzzes were used as indicators of prey capture attempts.

2. Statement of Naval Relevance

The results of this study provide important information about the biology and behavior of federally protected sperm whales. Due to the sparse amount of data available on sperm whales in the middle to southern U.S. Atlantic coastal region, this work is important to better understand the distribution, foraging ecology, and habitat preference of sperm whales in this region. This information will also help the Atlantic Operational Navy to meet the environmental stewardship obligations of the National Environmental Policy Act, MMPA, ESA, and other related environmental legislation. Comprehensive management of the living marine resources in this region requires reliable and up-to-date information about the occurrence, behaviors, and ecology of species inhabiting these areas. The results of this study will improve our understanding of occurrence, habitat use, and acoustic behaviors of sperm whales in Atlantic Operational Navy regions.

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3. Methods

3.1 Data Collection and Event Selection

Archival passive acoustic Navy funded datasets recorded off the coast of Jacksonville, Florida, Onslow Bay, North Carolina, and Cape Hatteras, North Carolina, were analyzed in this study (**Figure 1**). The dataset used for this study consisted of acoustic data from ten autonomous recorders; nine MARU's and one Autonomous Multi-channel Acoustic Recorder (AMAR).

3.1.1 Jacksonville

In 2009, nine MARUs were deployed in the U.S. Navy's planned Undersea Warfare Training Range (USWTR), which is located approximately 60 to 150 kilometers (km) offshore of Jacksonville, Florida. Three MARUs were deployed in shallow-water sites (44 to 47 meters [m]), three in mid-water sites (160 to 205 m), and three in deep-water sites (>300 m). These MARUs recorded continuously for 23 days in fall (JAX-1: 12 September to 4 October) and 34 days in winter (JAX-2: 4 to 26 December; **Table 1**). Because sperm whale vocalizations were detected only on the mid-depth recorders, (Norris et al. 2013) only data from these recorders were used in the current analysis.

3.1.2 Onslow Bay

Five MARUs were deployed in Onslow Bay during summer 2008. Each unit sampled continuously at 32 kHz for 23 recording days from 5 July to 27 July 2008 (**Table 1**). Two MARUs were deployed in shallow water (64 to 73 m), one in mid-depth water (236 m), and two in deeper water (approximately 366 m). Data from these recorders were initially examined for sperm whale clicks by Hodge et al. (2013), which showed that sperm whale vocalizations were only present at mid-depth and deep recorders. Bio-Waves, Inc. used the acoustic logs produced by Hodge et al. (2013) as a guideline to identify sperm whale encounters for the analysis presented here.

3.1.3 Cape Hatteras

Four AMARs were deployed off Cape Hatteras for 34 days during winter 2013 (Martin et al. 2015). The AMARs recorded continuously with a sampling rate of 128 kHz from 16 November to 19 December 2013. Three recorders were deployed in an equilateral triangle at 1 km distance from each other with a fourth recorder located in the center of the triangle. The recorders were deployed at depths that varied between 427 and 626 m. Because the recorders were deployed within relatively close proximity to each other, we assumed that most click events were detected on all recorders. Therefore, acoustic data from only the deepest (626 m) deployed AMAR was used in this analysis (**Table 1**).

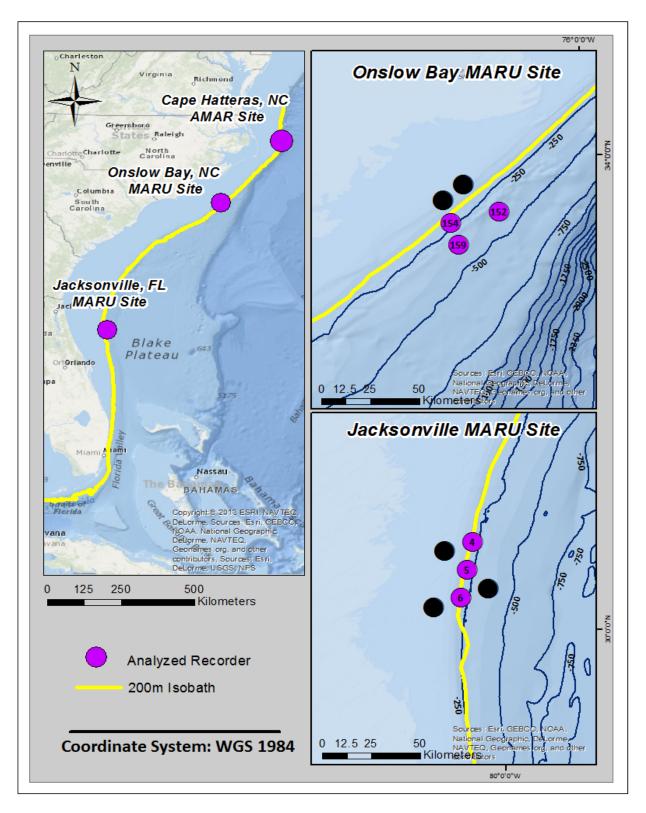


Figure 1. Recording regions reviewed for sperm whale vocal behavior. Three coastal U.S. Atlantic sites include Jacksonville, Florida, Onslow Bay, North Carolina, and Cape Hatteras, North Carolina. The Cape Hatteras site only had 1 recorder deployed at 626 m. Zoomed in regional maps for Onslow Bay and Jacksonville MARU maps show multiple recorder locations with bathymetry contour lines. The yellow line depicts the 200m isobath.

Table 1. Deployment information for MARUs (JAX and OB) sampled at 32 kHz and AMARs (HAT) sampled at 128 kHz used to collect the data supplied for analysis.

Region	Recorder/Site	Latitude (N)	Longitude (W)	Recorder Depth (m)	Deployment	Recording Start	Recording End	No. Recording Days
JAX-1	2 (Site 4)	30° 21.435′	80° 09.331′	168	1	12-Sep-09	4-Oct-09	23
JAX-1	74 (Site 5)	30° 14.505′	80° 10.879′	201	1	12-Sep-09	4-Oct-09	23
JAX-1	96 (Site 6)	30° 07.594′	80° 12.486′	192	1	12-Sep-09	4-Oct-09	23
JAX-2	2 (Site 4)	30° 21.357′	80° 09.170′	168	2	4-Dec-09	26-Dec-09	23
JAX-2	74 (Site 5)	30° 14.480′	80° 10.843′	201	2	4-Dec-09	26-Dec-09	23
JAX-2	96 (Site 6)	30° 07.609′	80° 12.503′	192	2	4-Dec-09	26-Dec-09	23
ОВ	PU152 (Site 1)	33° 43.546′	76° 22.132′	365	-	5-Jul-08	27-Jul-08	23
ОВ	PU154 (Site 2)	33° 40.454′	76° 35.382′	236	-	5-Jul-08	27-Jul-08	23
ОВ	PU159 (Site 3)	33° 34.164′	76° 33.309′	365	-	5-Jul-08	27-Jul-08	23
HAT	A3 (Site 1)	35° 45.414′	74° 49.080′	626	-	16-Nov-13	19-Dec-13	34

3.2 Logging Sperm Whale Encounters and Marking Click Events

3.2.1 Triton Logging

In order to prepare the acoustic data for analysis, all .wav (waveform audio file format) files were used to create long-term spectral averages (LTSAs) for each autonomous recorder using the MATLAB-based program Triton (Wiggins 2007). These LTSAs were then used to log (i.e., annotate) sperm whale acoustic encounters in all of the datasets.

Trained analysts scrolled through the LTSA for each autonomous recorder in Triton using a 30-minute resolution to identify sperm whale acoustic encounter periods. Encounters were defined as continuous periods of time containing sperm whale echolocation clicks with no more than a 30-minute interval between echolocation clicks. When more than 30 minutes occurred between echolocation clicks, a new encounter was logged.

3.2.2 Identification of Echolocation Clicks and Foraging Buzz Events

All .wav files containing sperm whale encounters were post-processed using an automated echolocation click detector in PAMGuard (Gillespie et al. 2008). The output of this processing was a database of binary files, which were used for additional post-processing. The detector was optimized to maximize the ratio of true positive clicks to false detections. Once all encounters were processed using PAMGuard, each encounter was further processed in PAMGuard's Viewer Mode software to mark click train and foraging buzz events in the dataset Analysts scrolled through each encounter using a 30-minute window to identify and select click trains, which were defined as any series of three or more consecutive clicks occurring within 2 minutes or less. If the interval between clicks was greater than two minutes, it was considered a new click train event (Figure 2). While selecting click trains, analysts spot-checked the clicks to make sure that they contained only true positive detections, by evaluating the waveform. frequency spectrum, and Wigner-Ville plot of individual clicks (Figure 2). Analysts also referred to the spectrogram to confirm periods of clicks. Once each click train in an event was identified and selected, it was examined at a finer timescale (e.g., < 1 minute) to identify and mark foraging buzz events (i.e., single buzzes) in the data. After identifying all click trains and foraging buzzes in an encounter (Figure 2), clicks were exported to the Real-time Odontocete Call Classification Algorithm (ROCCA), a module in PAMGuard, which was used to obtain click counts, echolocation click measurements, and detailed time and duration information (Appendix A).

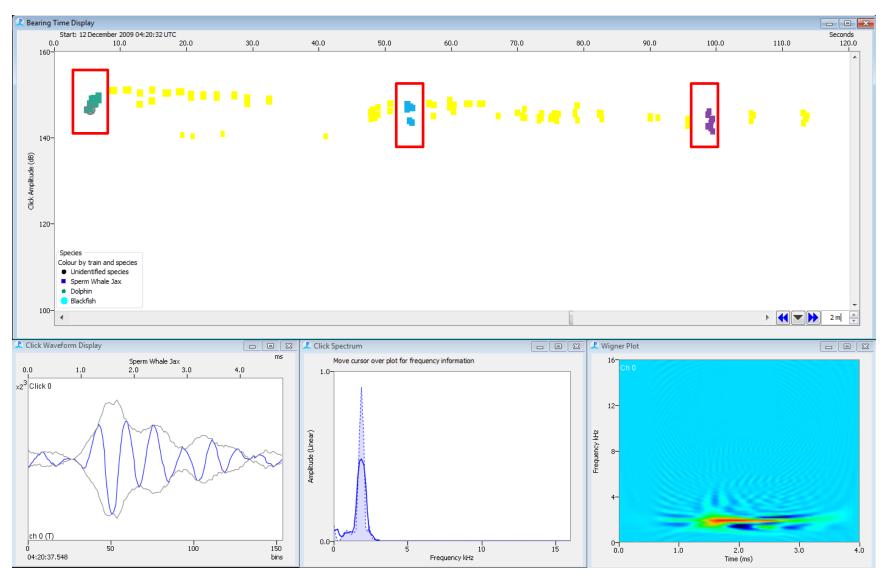


Figure 2. Example of click train selection in PAMGuard Viewer Mode click analysis. The top window shows the bearing time display, where click trains are selected. Clicks marked in different colors in the bearing time display are different click events. Yellow clicks represent a click train event, and clicks in red boxes represent foraging buzz events. The lower three windows (left to right) show the click waveform, click spectrum, and Wigner-Ville plot of individual clicks.

3.3 Data Analysis

Statistical analyses and plots were produced in RStudio Version 0.99.486 using R Version 3.2.2 (R Core Team 2015). Packages used during data analysis include maptools (Bivand and Lewin-Koh 2016), reshape2 (Wickham 2007), ggplot2 (Wickham 2009; used to produce all plots), and dunn.test (Dinno 2016).

3.3.1 Vocal Behavior

Sperm whale occurrence was plotted from the encounter logs by day and also by time of day for each recorder to provide an overview of vocal activity for each recording instrument. Photoperiod (i.e., day versus night) was assigned to each day of the plot using 'sun-methods' from the 'maptools' package (which uses algorithms provided by the National Oceanic and Atmospheric Administration [NOAA]; http://www.inside-r.org/packages/cran/maptools/docs/crepuscule). For each recorder, we calculated the number of click trains, the number of foraging buzzes, the number of days with click trains per day (number of click trains/number of recording days), the total duration of click trains (determined by measuring the time between the start and end time of a click train), the total duration of foraging buzzes, the percent of days with click trains, the percent of days with foraging buzzes (number of days with click trains or foraging buzzes/number of recording days), the proportion of time vocally active (sum of duration of click events/total recording duration), and the proportion of vocalization time spent foraging (duration of foraging buzzing/sum of duration of all click train events).

The G-test (or likelihood ratio test) is an alternative to Pearson's chi-square test (goodness of fit) for which the test statistic is the ratio of the probability of the observed frequencies of a variable to the probability of the expected frequencies:

$$G = 2 x \sum_{all \ cells} [Observed \ x \ \ln \left(\frac{Observed}{Expected} \right)]$$

The test statistic, G, is distributed as a chi-square variable with the degrees of freedom calculated the same as for Pearson's chi-square test. Expected frequencies can be calculated for each cell in a contingency table by dividing the product of that cell's row total and column total by the total sample size. The G-test was chosen for the following analysis because it was designed for field-type studies (where the row and column totals for variables are not fixed *a priori*) and can be more robust to small sample sizes than Pearson's chi-square test (Gotelli and Ellison 2013).

G-tests were performed on contingency tables of the following variables to test the null hypothesis that each of the following variables is independent of region/deployment (HAT, JAX_1, JAX_2, and OB): 1) number of days with (and without) clicks, 2) number of days with (and without) buzzes, 3) number of vocalizations that are regular clicks (versus foraging buzzes), and 4) the number of seconds spent clicking (versus buzzing). The JAX deployments (JAX_1 and JAX_2) were examined in more detail by executing the G-test on the same variables by only these two deployments. The multiple comparisons Dunn's test with Bonferroni

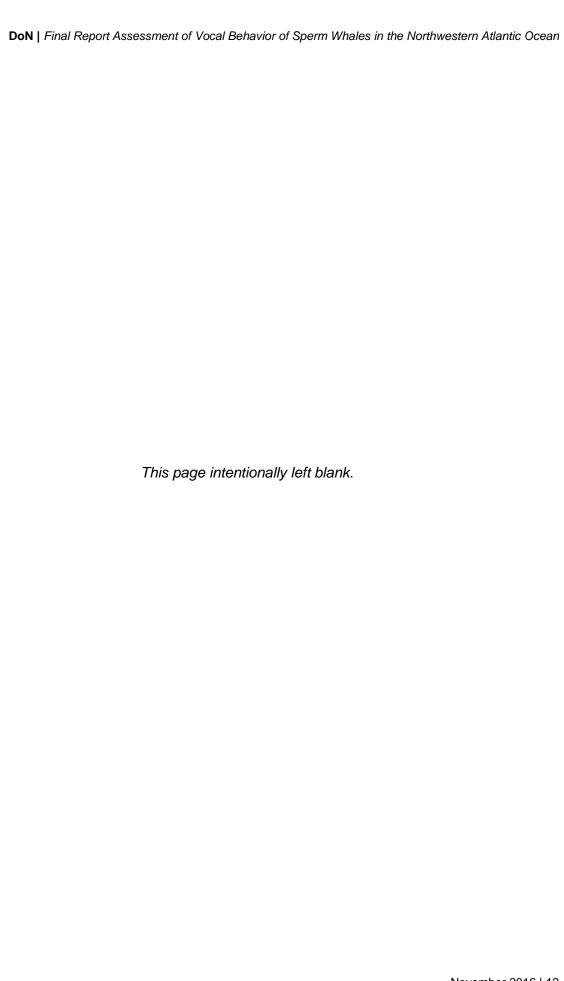
correction (in the dunn.test package) along with the associated overall Kruskal-Wallis test (Zar 1999) was used to test for regional differences in the number of clicks and buzzes per day.

3.3.2 Diel Pattern Analysis

We examined diel patterns in the occurrence of vocal events by dividing the recordings into 3-hour and 1-hour time bins. The number of clicks in each time bin was then calculated for each day using custom MATLAB code 'Bin-It Counter'. Photoperiod (day versus night) was assigned to each three-hour time bin to broadly categorize bins as either light or dark as follows: 00:00–03:00, 03:00–06:00, 18:00–21:00, and 21:00–24:00 were designated as "dark" time bins, the other four bins were designated as "light" time bins. For each calendar date, we summed the number of clicks within each photoperiod (light versus dark). The data violated the parametric assumption that modelled residuals conform to a normal distribution, so Kruskal-Wallis tests were used to determine whether – for each site and overall – there were any differences in the number of clicks between: (1) photoperiods, (2) 3-hour time bins, and (3) hourly time bins. Differences between regions in the number of clicks in each photoperiod were also tested with a Kruskal-Wallis test and explored further by performing Dunn's tests with Bonferroni corrections to test pairwise site differences.

3.3.3 Click Analysis

Echolocation clicks from click train and foraging buzz events were measured using PAMGuard ViewerMode and new tools developed for the PAMGuard Module ROCCA (**Appendix A**). Measurements included duration (microseconds), center and peak frequency (kHz), number of zero crossings, sweep rate (kHz/millisecond), and ICI (second). A Kruskal-Wallis test was used to determine if there was significant variability in the measures among sites, and a multiple comparison Dunn's test with Bonferroni correction was performed to determine significant differences. Because the distributions of these vocalization measures by site did not always have the same shape and scale, results of randomization tests (with 10,000 replicates) of the ANOVA F-statistic by site and the pairwise median measure differences between sites were compared to the results of the Kruskal-Wallis and Dunn's tests, respectively. The latter randomization test was based on the MED procedure described by Richter and McCann (2013), in which each pairwise median difference is compared to the distribution of the maximum pairwise median difference generated in each randomization replicate.



4. Results

4.1 Vocal Behavior

Data were analyzed from 10 different bottom-mounted recorders from three different geographic regions. Vocal behavior varied both within and among the regions/deployments compared. Overall the highest number of encounters and highest total duration of encounters occurred during JAX -1 (*n*=90 encounters, approximately 113 hours [hr]), followed by JAX-2 (*n*=46 encounters, approximately 68 hr), OB (*n*=39 encounters approximately 50 hr), and HAT (*n*=28 encounters, approximately 23.5 hr). A total of 4,108 click train events and 43 foraging buzz events were identified (**Table 2**).

The proportion of days with click trains and foraging buzzes present was highest overall at the JAX sites, followed by HAT and OB, respectively (**Table 2**). The number of click train events per day was highest at JAX-1 (22), followed by OB (18), JAX-2 (17), and HAT (5) (**Table 2**). The number of foraging buzzes per day was an order of magnitude lower relative to click train events, but relative to recorder site was highest overall at JAX-2 (0.36), followed by JAX-1(0.23), HAT (0.03), and OB (0.01) (**Table 2**).

Table 2. Summary of sperm whale vocal activity by site and region/deployment.

Site ID	Recorder ID	Recorder depth (m)	# Recording Days		# Foraging Buzz Events	# Clicks	# Days with Click Train Events	# Days with Foraging Buzz Events	Click Trains per Day	Foraging Buzzes per Day	Proportion of Days with Click Trains	Proportion of Days with Foraging Buzzes	Proportion of Vocalization Time in Prey Capture Attempt
JAX-1-4	Site 4 (PU2)	167.6	23	730	3	71,859	19	3	32	0.13	83%	13%	0.380%
JAX-1-5	Site 5 (PU74)	201.5	23	301	0	17,237	9	0	13	0	39%	0%	0.000%
JAX-1-6	Site 6 (PU96)	191.7	23	517	13	19,207	16	4	22	0.57	70%	17%	0.053%
	JAX-1 Total		69	1,548	16	108,303	44	7	22	0.23	64%	10%	0.207%
JAX-2-4	Site 4 (PU2)	167.6	23	170	19	14,264	11	5	7	0.83	48%	22%	0.052%
JAX-2-5	Site 5 (PU74)	201.5	23	172	4	8,722	10	1	7	0.17	43%	4%	0.027%
JAX-2-6	Site 6 (PU96)	191.7	23	811	2	78,953	23	1	35	0.09	100%	4%	0.041%
	JAX-2 Total		69	1,153	25	101,939	44	7	17	0.36	64%	10%	0.041%
HAT-1-1	A3	558	34	182	1	43,277	14	1	5	0.03	41%	3%	0.001%
	HAT Total		34	182	1	43,277	14	1	5	0.03	41%	3%	0.001%
OB-1-1	Site SB3 (PU152)	365.8	23	10	0	110	1	0	0	0	4%	0%	0.000%
OB-1-2	Site DB2 (PU154)	236.2	23	1169	1	70,891	21	1	51	0.04	91%	4%	0.001%
OB-1-3	Site SB5 (PU159)	365.8	23	46	0	1,332	3	0	2	0	13%	0%	0.000%
	OB Total		69	1,225	1	72,333	25	1	18	0.01	36%	1%	0.001%

The percentage of recording days with vocal activity varied by recording site and geographic region (**Table 2, Figures 3 through 5**). Click trains were present every day of the JAX-2-6 recorder deployment, 91 percent of days at OB site 1-2 and 83 percent of days at JAX-1-4 (**Table 2, Figure 3**). Foraging buzzes were present during the highest percentage of days overall during both JAX deployments (10 percent) followed by HAT (3 percent), and OB (1 percent) (**Table 2, Figure 3**).

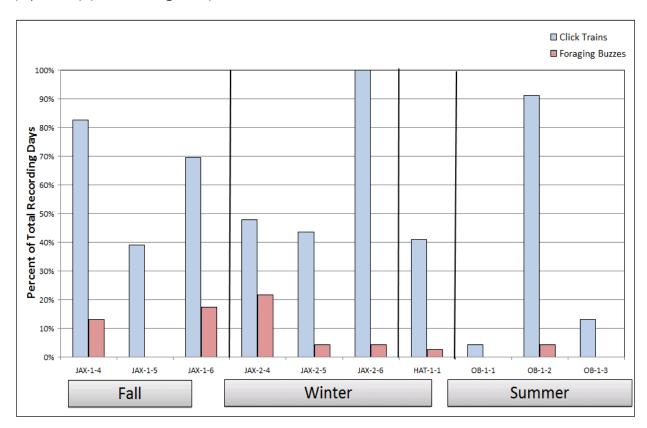


Figure 3. Percentage of recording days (y-axis) with click trains (blue) and foraging buzzes (rose) present at each site (x-axis). The season that the recording occurred in is denoted beneath the x-axis.

Daily click train and foraging buzz rates varied by site and region (**Table 2, Figure 4**). OB site 1-2 had the highest occurrence of click trains per day (51) but a low occurrence of foraging buzzes per day (0.04; **Figure 4**). The highest number of foraging buzzes per day occurred at JAX-2-4 (0.83; **Figure 4**).

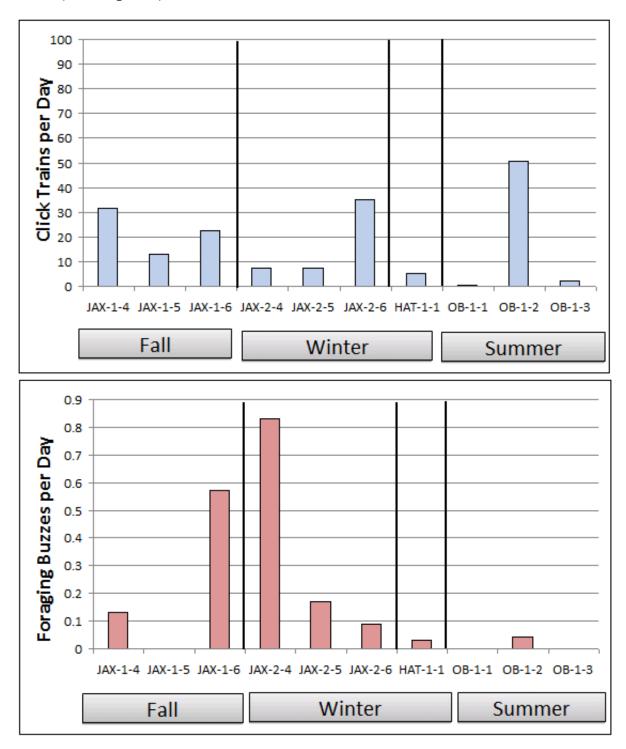


Figure 4. The total number of click trains (top) and foraging buzzes (bottom) per day (y-axis) at each recording site (x-axis). The season that the recording occurred in is denoted beneath the x-axis. Note the order of magnitude difference in scale of the Y-axis between the top and bottom graphs.

Sperm whales were vocally active for the greatest percent of total recording time at JAX-1-4 (11 percent), followed by JAX-2-6 (10 percent) and OB site 1-2 (9 percent), respectively (**Figure 5**).

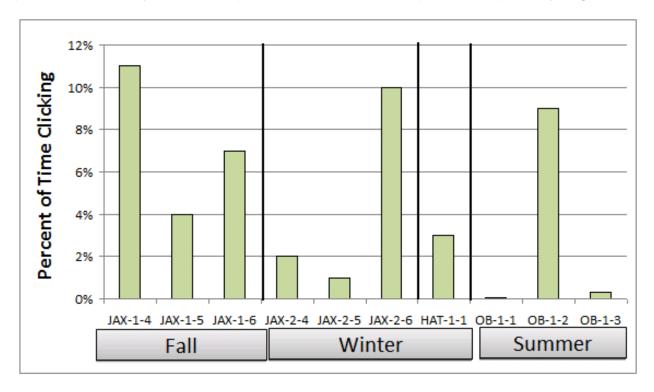


Figure 5. The percentage of total recording time (y-axis) that sperm whale clicks were detected at each recording site (x-axis). The season that the recording occurred in is denoted beneath the x-axis.

The number of regular clicks per day was plotted as a function of recorder depth (**Figure 6a**) and distance from the 200-m isobaths (**Figure 6b**). There was no recognizable relationship between clicks per day and depth; however, it appears that fewer clicks per day were detected on recorders located greater than 10 km from the 200-m isobath.

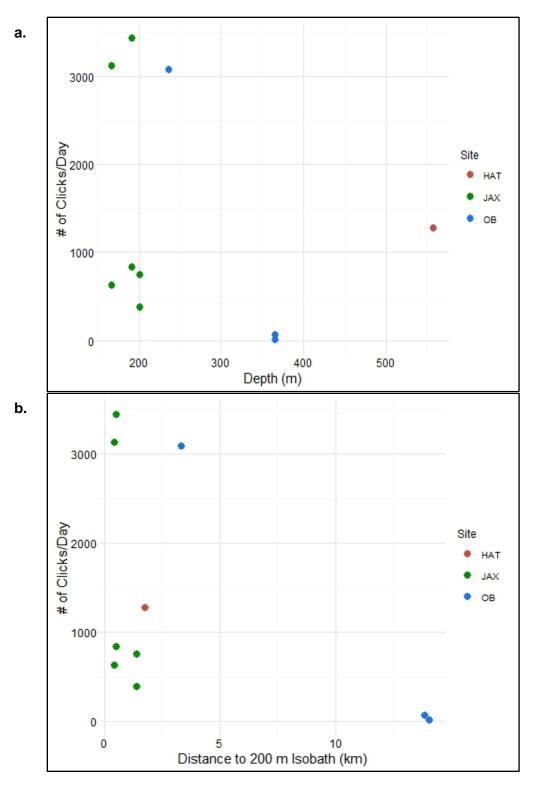


Figure 6. Scatter plots showing the number of clicks per day as a function of recorder depth (top) and recorder distance from the 200-m isobath (bottom). Study region is represented by color; red (HAT), green (JAX), and blue (OB). Sperm whale vocal activity was plotted by day and time for each recording site in each region (Figures 7 through 10).

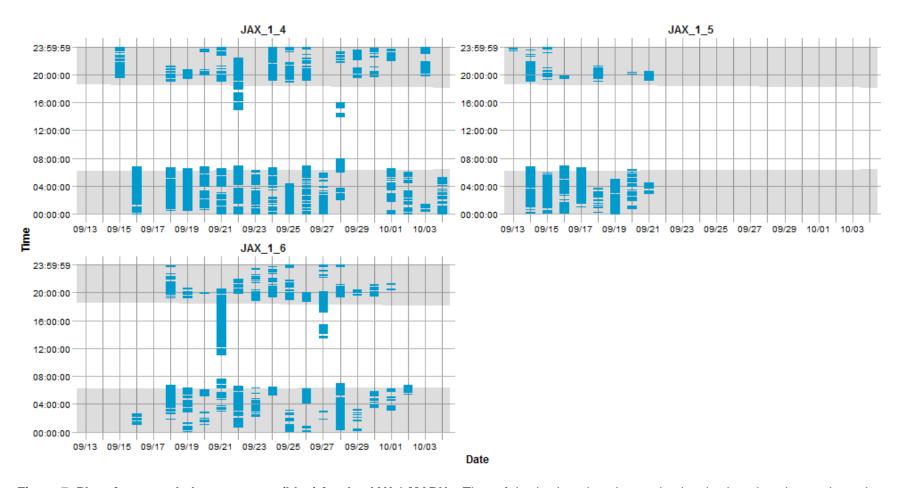


Figure 7. Plot of sperm whale encounters (blue) for the JAX-1 MARUs. Time of day is plotted on the y-axis, date is plotted on the x-axis, and shading represents periods of light and darkness.

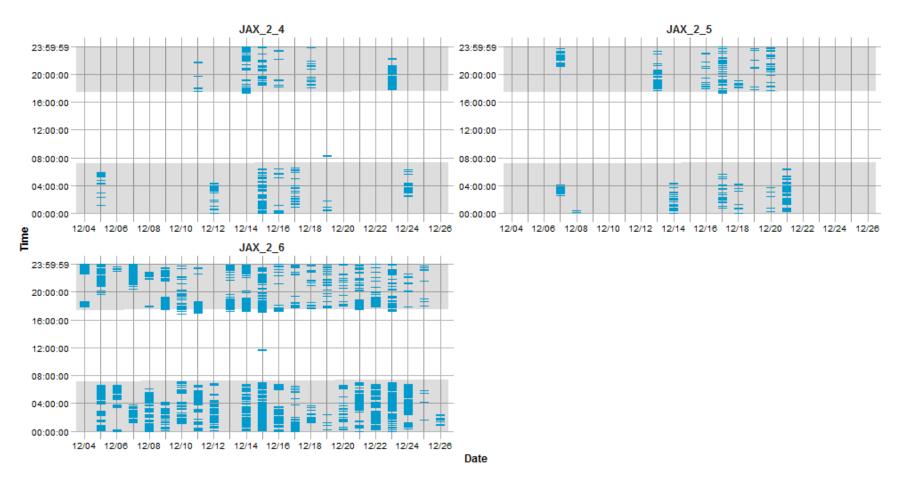


Figure 8. Plot of sperm whale encounters (blue) for the JAX-2 MARUs. Time of day is plotted on the y-axis, date is plotted on the x-axis, and shading represents periods of light and darkness.

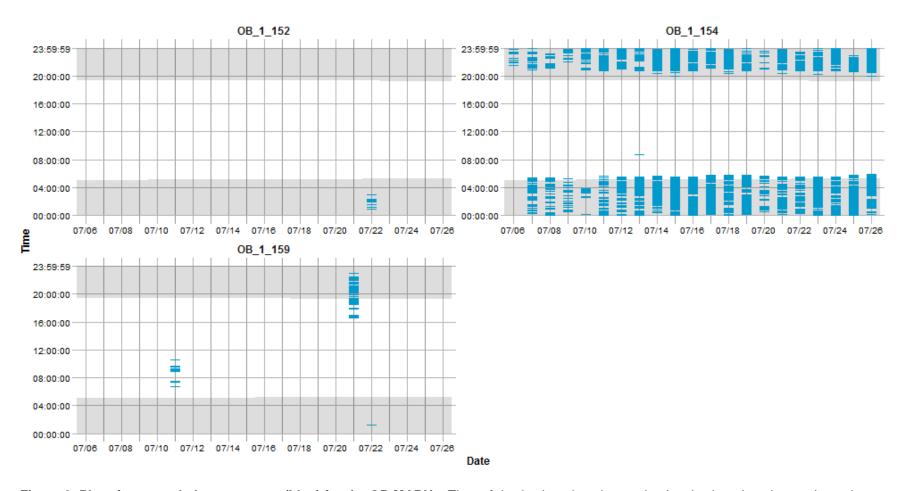


Figure 9. Plot of sperm whale encounters (blue) for the OB MARUs. Time of day is plotted on the y-axis, date is plotted on the x-axis, and shading represents photoperiods (light and dark).

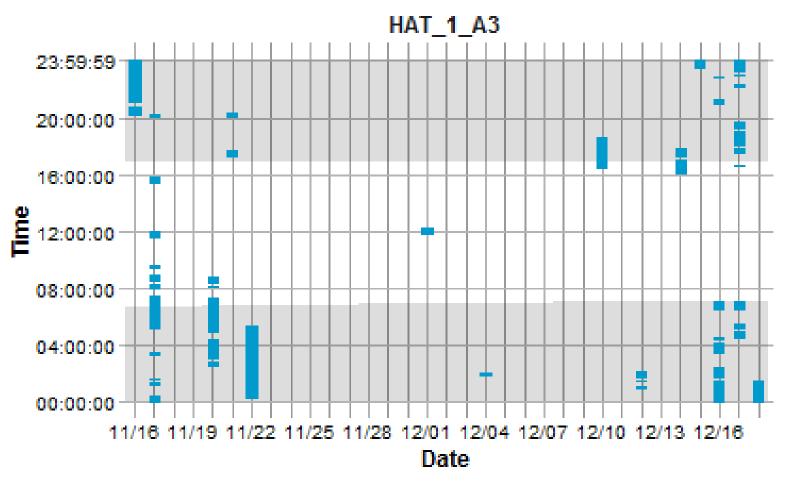


Figure 10. Plot of sperm whale encounters (blue) for the HAT AMARs. Time of day is plotted on the y-axis, date is plotted on the x-axis, and shading represents photoperiods (light and dark).

4.1.1 Statistical Analysis of Vocal Behavior

The proportion of days with clicks was significantly different (p< 0.05) from that expected among all regions if it were independent of region, but the null hypothesis could not be rejected when looking at only the two JAX deployments (i.e., fall versus winter; **Table 3**). The proportion of days with foraging buzzes was not significantly different from that expected given independence among regions or between the two JAX deployments (**Table 3**). The total number of clicks compared to the total number of foraging buzzes was shown to be dependent on all regions and the two JAX deployments, as was the number of seconds spent regular clicking versus buzzing (**Table 3**).

Table 3. P-values resulting from G-Tests to test for significant deviations (red) from the null hypothesis of independent associations among all region deployments and between the two JAX deployments (fall versus winter).

Regional G-Test								
(H ₀ = "Variable" and "Region" (or "JAX Deployment") are Independent)								
Variable p{Region} p{JAX Deployments Seasonal Comp								
Proportion of Days w/ Clicks	<0.05	1.00						
Proportion of Days w/ Foraging Buzzes	0.054	1.00						
Ratio of Clicks to Foraging Buzzes	<0.05	<0.05						
Ratio of Duration of Clicking vs. Foraging Buzzing	<0.05	<0.05						

The average daily regular click rate was significantly different overall among deployments/ regions and between HAT and all deployments but not between OB and the JAX deployments or between the two JAX deployments (**Table 4**). The average daily foraging buzz rate was significantly different overall among deployments/regions and between HAT and the JAX-2 deployment but not between HAT and JAX-1, OB and the two JAX deployments or HAT deployments, or between the two JAX deployments (**Table 4**).

Table 4. P-values resulting from Kruskal-Wallis test to assess significant variability among region/deployment and Dunn's test with Bonferroni correction to test for pairwise significant differences (red) in the regular click and foraging buzz rates.

Vocal Rate	Region	Dunn's	Test Resul	Kruskal-Wallis Test Results	
Vocal Rate		HAT	JAX-1	JAX-2	Overall
	JAX-1	<0.05	-	-	
Regular Clicks/Day	JAX-2	<0.05	0.62	-	<0.05
Gileite, Bay	ОВ	<0.05	1.00	1.00	
	JAX-1	0.08	-	-	
Foraging Buzzes/Day	JAX-2	<0.05	1.00	-	<0.05
242200, 24y	ОВ	1.00	0.20	0.06	

4.2 Diel Patterns

The occurrence plots (**Figures 7 through 10**) and plots of click counts summed in hourly bins within each geographic region (**Figure 11**) suggest that there are strong diel patterns of sperm whale vocal activity in the JAX and OB regions.

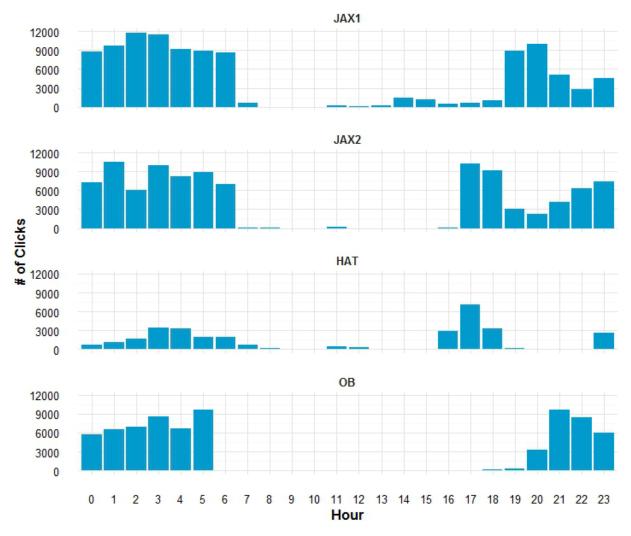


Figure 11. Histograms of click counts (y-axis) in hourly bins (x-axis) in each recording region/deployment.

To examine these apparent patterns in more detail we used a Kruskal-Wallis test to determine if there were significant differences in the number of clicks between: (1) 3-hour time bins within and among sites, (2) photoperiods (light versus dark) within and among sites and (3) hourly time bins within and among sites. Click counts were found to be significantly different among 3-hour time bins, 1-hour time bins and between photoperiods at every site except HAT and were significantly different among all sites overall (**Table 5**).

Table 5. P-values resulting from Kruskal-Wallis tests for significant variability(red) in click counts between photoperiods (light and dark) and among 3-hour time bins and 1-hour time bins by region/deployment and overall.

Toot Groups	Region/Deployment								
Test Groups	HAT	JAX-1	JAX-2	ОВ	Overall				
Photoperiod	0.10	<0.05	<0.05	<0.05	<0.05				
3-hr Time Bins	0.34	< 0.05	< 0.05	< 0.05	<0.05				
1-hr Time Bins	0.12	<0.05	<0.05	< 0.05	<0.05				

Multiple comparison Dunn's tests with Bonferroni corrections were also performed to determine how diel patterns varied within sites to assess whether there were significant differences in click counts within photoperiods among sites.

There were only significant differences in click counts within light periods at OB compared to all other sites and overall among regions (**Table 6**). There were only significant differences within dark periods at HAT compared to all other sites and overall among sites (**Table 6**).

Table 6. P-values resulting from Kruskal-Wallis test to assess significant variability among region/deployment by photoperiod and Dunn's test with Bonferroni correction to test for significant pair-wise differences (red) between region/deployment.

Dheteneried	Dogion	Dunr	's Test Re	sults	Kruskal-Wallis Test Results		
Photoperiod	Region	HAT	JAX-1	JAX-2	Overall		
	JAX-1	1.00	-	-			
Light	JAX-2	0.15	0.27	-	<0.05		
	ОВ	<0.05	<0.05	<0.05			
	JAX-1	< 0.05	-	-			
Dark	JAX-2	<0.05	0.23	-	<0.05		
	ОВ	<0.05	0.86	1.00			

4.3 Echolocation Click Feature Analysis

Box plots of click measurements, combined by geographic region, were used to assess whether there are regional differences in the features of regular clicks (**Figure 12**, **Table 7**). The sample size of foraging buzzes was too low to compare among regions.

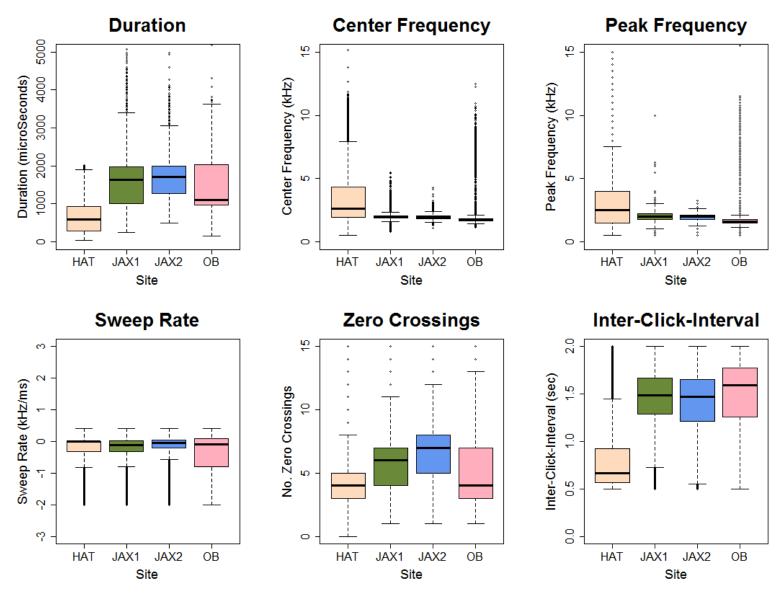


Figure 12. Box plots displaying the regular click paramaters measured from each region/ deployment. The boxes represent the upper 75 percent quartile and the lower 25 percent quartile, with the solid black horizontal line indicating the median. The hinges show the maximum and minimum values and the stars represent outliers that are more than or less than 1.5 times the quartile ranges, respectively. Region (HAT, JAX-1, JAX-2, and OB) is along the x-axis and each parameter is along the y-axis.

Table 7. Medians and 10th–90th percentile ranges (in parentheses) for variables measured from regular echolocation clicks by region/deployment.

Region	Peak Frequency (kHz)	Center Frequency (kHz)	Duration (μs)	Sweep Rate (kHz/ms)	ICI (sec)	No. Zero Crossings	BW 3 dB (kHz)	BW 10 dB (kHz)	N
НАТ	2.00	2.45	562.5	0.00	0.67	2	2.00	3.50	5,689
ПАТ	(1.50–3.00)	(1.91–3.69)	(281.0–914.1)	(-0.50-0.00)	(0.57-0.91)	(2-4)	(1.50–2.50)	(2.50-5.00)	
JAX-1	2.00	1.98	1,688.0	-0.15	1.49	7	0.75	1.50	36,497
JAX-1	(1.75–2.12)	(1.88–2.07)	(1,125.0-2,000.0)	(-0.37-0.00)	(1.30-1.67)	(5–8)	(0.7–1.00)	(1.2–1.75)	
JAX-2	2.00	1.95	1,781.0	-0.12	1.46	7	0.75	1.38	33,841
JAX-2	(1.75–2.00)	(1.85–2.06)	(1,531.0-2,031.0)	(-0.24-0.00)	(1.20-1.65)	(6–8)	(0.75–1.00)	(1.25–1.50)	
ОВ	1.63	1.73	1,094.0	-0.18	1.60	3	0.75	1.50	10 005
OB	(1.50–1.75)	(1.66–1.81)	(968.8–2,031.0)	(-0.89-0.05)	(1.32-1.78)	(3–7)	(0.5–1.00)	(0.8–2.00)	18,235

^{*}BW = bandwidth

Kruskal-Wallis test results showed significant variability among deployments for all six of the regular echolocation click parameters (**Table 8**). The Dunn's test results showed statistically significant pair-wise difference between deployments for all of the regular echolocation click parameters, except for peak frequency between the JAX-2 deployment and HAT (**Table 8**).

Table 8. P-values resulting from Dunn's tests with Bonferroni corrections for significant pair-wise differences (red) between sites, and Kruskal-Wallis tests for significant variability among sites, for each regular click measure.

Regular Click	Region	Du	nn's Test Resu	Kruskal-Wallis Test	
Measure	Region	HAT	JAX-1	JAX-2	Results
Peak Frequency	JAX-1	<0.05	-	-	<0.05
	JAX-2	0.12	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
Center Frequency	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
Duration	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
Sweep Rate	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
ICI	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
# Zero Crossings	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	

We also performed randomization tests to better assess pairwise comparisons and differences overall among regions, as well as to compare these results to the Dunn's test results. The among-region comparisons showed that all regular click measure parameters were significantly different (**Table 9**). However, pairwise comparisons showed slightly fewer significant differences than the results of the Dunn's test. Peak frequency was shown to be significant only at Onslow Bay compared to all other deployments and the number of zero crossings was not significantly different between the two Jacksonville deployments (**Table 9**).

Table 9. Regular click measure p-values resulting from randomization tests where p is the proportion of permutated statistics greater than or equal to the test statistic (red indicates

significant differences at $\alpha = 0.05$).

Click Measure	Region/Deployment	HAT	JAX-1	JAX-2	Overall
Peak Frequency	JAX-1	1.00	-	-	<0.05
	JAX-2	1.00	1.00	-	
	ОВ	<0.05	<0.05	<0.05	
Center Frequency	JAX-1	< 0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	< 0.05	<0.05	< 0.05	
Duration	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
Sweep Rate	JAX-1	< 0.05	-	-	<0.05
	JAX-2	<0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
ICI	JAX-1	<0.05	-	-	<0.05
	JAX-2	< 0.05	<0.05	-	
	ОВ	<0.05	<0.05	<0.05	
# Zero Crossings	JAX-1	<0.05	-	-	<0.05
	JAX-2	<0.05	1.00	-	
	ОВ	<0.05	<0.05	<0.05	

Note: For pairwise site differences, the test statistic is the median difference in the click measure compared to a distribution of permutated maximum median differences. To test differences over all sites ('Overall'), the ANOVA F-statistic was compared to a distribution of permutated F-statistics.

4.4 Relationship to Historical Visual Data

Sighting and tag data were obtained from OBIS-SEAMAP (Ocean Biogeographic Information System Spatial Ecological Analysis of Megavertebrate Populations; Halpin et al. 2009) to compare with acoustic recorder sites and plotted by type of observations (**Figure 13**). Historical sighting data show the majority of sperm whale visual detections occur in the northern Atlantic regions, north of Cape Hatteras, North Carolina, as well as beyond the continental slope. Few visual observations of sperm whales were found historically within and around the Jacksonville, Florida, and Onslow Bay, North Carolina sites. Comparatively, visual observations of sperm whales adjacent to Cape Hatteras, North Carolina, are more abundant. Historical sightings show sperm whales inhabiting the northern and southern Atlantic waters during all seasons, although sightings in fall and winter are more prevalent in the southern Atlantic waters. These data must be interpreted with caution, as search effort is not included in the map.

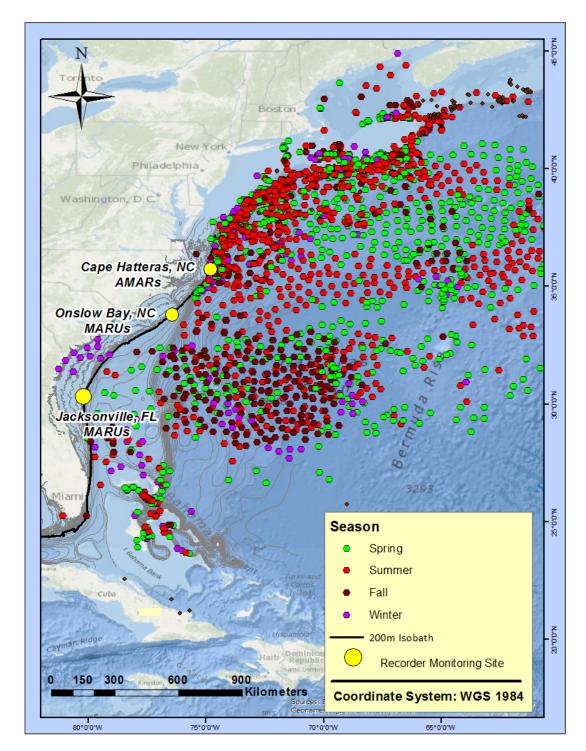


Figure 13. Map of sperm whale sighting data obtained from OBIS-SEAMAP in comparison to recorder locations. Data from OBIS-SEAMAP (Halpin et al. 2009) plotted by season (spring = green; summer = red; fall = brown; winter = purple). Recorder monitoring sites are indicated by yellow circles. Data retrieved from multiple datasets (Townsend 1935; Brown et al. 1975; CETAP 1982; Potter and NMFS 1991, 1995a, 1995b, 2002; SEFSC 1992, 1999; NEFSC 1995, 1997, 1998a, 1998b, 1998c, 2002, 2004, 2010, 2011a, 2011b, 2013a, 2013b, 2013c; NOAA 1998; UNCW 1999a, 1999b; Whitehead and Dalhousie University 2004, 2005; BMMRO 2006a, 2006b, 2006c; McLellan and UNCW 2006, 2008, 2010, 2012a, 2012b, 2013a, 2013b, 2014a, 2014b, 2014c; Jochens et al. 2008; HDR|EOC 2011; NEFSC and SEFSC 2011, 2013; Dunn 2012; Gatzke et al. 2013; IFAW et al. 2013; Kopelman 2013; DUML 2014).

5. Discussion

The interpretation and discussion of results from this analysis must be prefaced by acknowledging several caveats. First, there was limited spatial sampling at each of the three study regions. This was especially the case at Cape Hatteras, where the close spacing (1 km) among recorders only allowed analysis of one recorder at that site because recorders were not independent. Recorders at Jacksonville and Onslow Bay were deployed at shallow, mid-depth, and deep-water locations. In Onslow Bay, the mid-depth recorder was placed approximately 12 km and 21 km from the two deep-water recorders, and the deep-water recorders were placed approximately 24 km apart. In Jacksonville, the mid-depth recorders were spaced 15 km apart, and 15 km and 26 km from the deep-water recorders. Our results suggest that sperm whales in these regions preferentially forage at mid-depth locations (Norris et al. 2012, Hodge et al. 2013); however, spatial sampling was still limited longitudinally along the shelf break.

The second caveat is that there was temporal variation in sampling. Cape Hatteras was sampled for 34 days during the winter, Onslow Bay was sampled for 23 days in summer, and Jacksonville was sampled during two 23-day deployments in fall and winter. Therefore, seasonal comparisons could only be made for the Jacksonville deployments between the fall and winter, and any interpretation of the regional comparisons must be tempered by acknowledging that underlying seasonal variation could not be accounted for in the current analysis.

Thirdly, the total sample size of foraging buzzes was very low (N=43), with almost all foraging buzzes (95 percent) detected at the Jacksonville recorder sites. The low number of foraging buzzes detected could be due in part by the fact that foraging buzzes are highly directional and significantly lower in amplitude compared to regular clicks (Madsen et al. 2002, Goold and Jones 1995). Consequently, it follows that the detection ranges will be much shorter for buzzes than regular clicks. Because of the limited detection ranges for buzzes and the low sample size, the statistically significant differences in foraging buzz rates among and within recording sites must be interpreted with caution. Despite these caveats, we can say with certainty that feeding behavior occurred at all three study regions, albeit not at all recorder sites.

One final caveat that needs to be addressed is the possible relationship between detected vocal activity (clicks detected per day) at recorder sites and distance from the recorder to the 200-m isobath. Distance to the 200-m isobath appears to be loosely correlated with vocal activity, suggesting that at distances greater than 10 km from the 200-m isobath there are fewer sperm whale clicks detected per day. One possible interpretation of this result is that some recorders were positioned too far from the immediate area of foraging activity and thus were not able to record foraging buzzes. Additional information, such as tracking or tagging animals, would be needed to further investigate this possibility.

5.1 Regional Differences in Vocal Behavior

Our results indicate that relative detections of click trains were highest at JAX during both (fall and winter) deployments followed by OB (summer), both with respect to the percentage of days with click trains and foraging buzzes detected and the number of click trains and foraging

buzzes detected per day (total click trains/total days of recordings). The proportion of days with clicks was not significantly different between the two JAX deployments, suggesting that sperm whale vocal behavior did not vary between the recording deployments. However, the mean daily regular click rate was significantly different overall among deployments and regions and between HAT and all deployments, but not between the OB and JAX deployments. This suggests that the Onslow Bay and Jacksonville sites are more similar to one another with respect to vocal behavior than either of the regions compared to Cape Hatteras.

Sperm whales feed on a wide variety of squid species (Whitehead et al. 2003) while in some regions, notably New Zealand and the northern parts of the Pacific and Atlantic oceans (Kawakami 1980), fish are the predominant component of their diet (Berzin 1972, Clarke and Macleod 1976, Gosho et al. 1984, Martin and Clarke 1986, Rice 1989). The two most common squid in the North Atlantic are the longfin inshore (Loligo pealeii) of the family Loliginidal and the northern shortfin (Illex illecebrosus) of the family Ommastrephidal (Staudinger 2006). Sperm whales stomach content analysis has revealed that squid of the family Ommastrephidae are a primary part of the diet and Loliginidal are also fed on to a lesser degree (Clarke 1996). Squid distribution on the northwestern Atlantic shelf is temporally variable as squid move between inshore waters in spring and summer to offshore waters in fall and winter (Macy and Brodziak 2001). Additionally, annual squid migration occurs along the Canadian and the United States eastern shelf from Newfoundland to south of Cape Hatteras. Studies of the northern shortfin squid suggest that this species migrates off the continental shelf and southward in fall, returning to the shelf and migrating northward in the spring (Hendrickson 2004, Hendrickson and Holmes 2004). Longfin inshore squid also exhibit similar migrations, moving off the shelf into deeper water in autumn and returning to shallower shelf waters in spring (Cargnelli et al. 1999). The single Hatteras deployment occurred during winter when squid were likely to be migrating southward and located offshore of the shelf, while the OB deployment occurred during summer when squid would be inshore, but would be migrating northward. As such, the regional differences in sperm whale vocal behavior cannot be fully explained without also considering seasonal variations in vocal behavior and as well as seasonal variation in prey availability. To investigate these possibilities further will require better temporal and spatial sampling such as year-round recorder deployments in each of the study regions, and placement of recorders at similar depths and distances from the shelf break at each site.

5.2 Regional Differences in Diel Patterns

Results of the non-parametric Kruskal-Wallis tests for photoperiod (i.e., day versus night), and comparisons for 3-hour time bins and 1-hour time bins indicated that all areas except HAT had significant variability in each of these time categories. This indicates that differences in clicking (and presumably foraging) activity differed with respect to time of day and photoperiod at the OB and JAX study areas. Although there is no direct evidence, it is possible that these differences are related to changes in prey availability, distribution, and/or abundance. Past research on sperm whale diel behavior has produced varying results, perhaps based on regional, population-level differences (Whitehead 2003, Aoki et al. 2007, Pastavartou et al. 1989, Davis et al. 2007, Hodge 2011, Barlow and Taylor 2005, Merkins 2013). In the Gulf of Mexico, diel patterns in sperm whale acoustic detections were found to be different at each of three High-frequency Acoustic Recording Package (HARP) deployment sites. One site showed that

there was no significant diel pattern, a daytime foraging pattern was observed at the second site and at the third site nocturnal and anti-crepuscular patterns were observed (Merkins 2013). Additionally, there is some evidence of nocturnal foraging from tag data off Japan (Aoki et al. 2011).

The consistent diel pattern that was evident at all sites, except HAT, suggests a difference in either the foraging or search strategy for sperm whales along the shelf break. This is consistent with the fact that the distribution for at least two species of squid known to occur near the shelf break varies temporally (i.e., they move on a diurnal basis from demersal waters during the day to surface waters at night; Lange and Sissenwine 1983). One possible interpretation of our results is that sperm whales in the Jacksonville and Onslow Bay study regions are targeting the vertical migration of prey at night, which allows foraging at shallower mid-water depths at night when prey availability is relatively greater near the surface and thus, less expensive, energetically, to forage on. In Cape Hatteras, the MARU deployment period was during winter when squid in that region are located primarily offshore of the shelf break (Cargnelli et al. 1999). As such, sperm whales in the vicinity of the HAT recorder deployment may be opportunistically foraging on other types of prey (e.g., fish) at night, and foraging on squid in deeper offshore waters during the day.

5.3 Regional Differences in Click Measurements

The randomization tests showed that all regular click measure parameters were significantly different overall among regions. Peak frequency was significantly different (p<0.05) only between Onslow Bay and all other deployments, and the number of zero crossings was not significantly different between the two Jacksonville deployments. Echolocation click characteristics may change depending on the animal's orientation and distance to the hydrophone, while the animal's depth also may affect frequency spectral content and ICI (Thode et al. 2002). Additionally, propagation effects can result in significant distortions and other changes to the signal at distance over several hundreds of meters or more. This might have affected the results of this study. Furthermore, some odontocetes, including sperm whales, are capable of changing the source level and spectral content of their biosonar (Madsen et al. 2002; Madsen and Payne, 2004). Madsen et al. (2002) have shown that sperm whales can regulate the sound pressure levels of their clicks, and further suggest that it is sonar or feeding demands rather than available air volume that dictate acoustic output levels. Additionally, Madsen and Payne (2004) have shown that free-ranging false killer whales (Pseudorca crassidens) and Risso's dolphins (Grampus griseus) have a dynamic sound generator which allows varying of source level and centroid frequencies, thereby illustrating that biosonar is not simply a static high-powered system with fixed beams. As such, it is possible that differences in regular click features of sperm whales may be explained by differences in this species' diving behavior, acoustic behaviors, prey selection, and foraging behavior in the study regions.

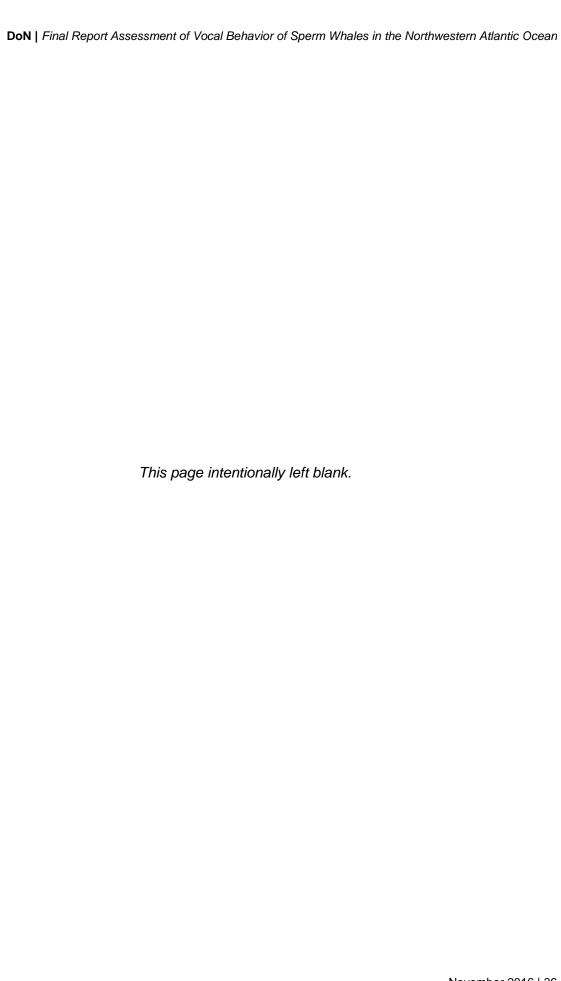
However, there are many factors that can contribute to the differences detected in click features in this study. Additional information with known distances, orientations, and age/sex-classes of sperm whales is necessary to better address the influence of these factors on the data.

This study provides new information about the distribution, occurrence, and vocal behaviors of sperm whales in the coastal northwestern Atlantic. Although there are limitations and caveats to

the interpretation of these data, the results here address gaps in current knowledge of sperm whale occurrence and behaviors, including the persistent presence, occurrence of foraging activity, and vocal behaviors of these deep-diving marine mammals in regions where they have been very rarely sighted using traditional visual methods. Additional sampling using both passive acoustic methods, such as towed-hydrophone-array surveys and tracking, coupled with electronic (e.g., satellite) tagging will be needed to provide more information about the occurrence and activities of sperm whales in the study regions. Research on the distribution, movement patterns, and vertical migration of the sperm whale's primary prey, squid, will also provide information to help interpret results. Our findings have identified that sperm whales are foraging primarily at night along the shelf break off Jacksonville and Onslow Bay. Additionally, this study has provided evidence of geographic variation in sperm whale vocal behavior among the study areas. The detection of click trains and foraging buzzes indicate that these areas may be important foraging habitat for sperm whales, for example off Jacksonville and Onslow Bay in particular. The results of this study provide important new information that can be used to better inform management, mitigation and conservation of this federally protected species.

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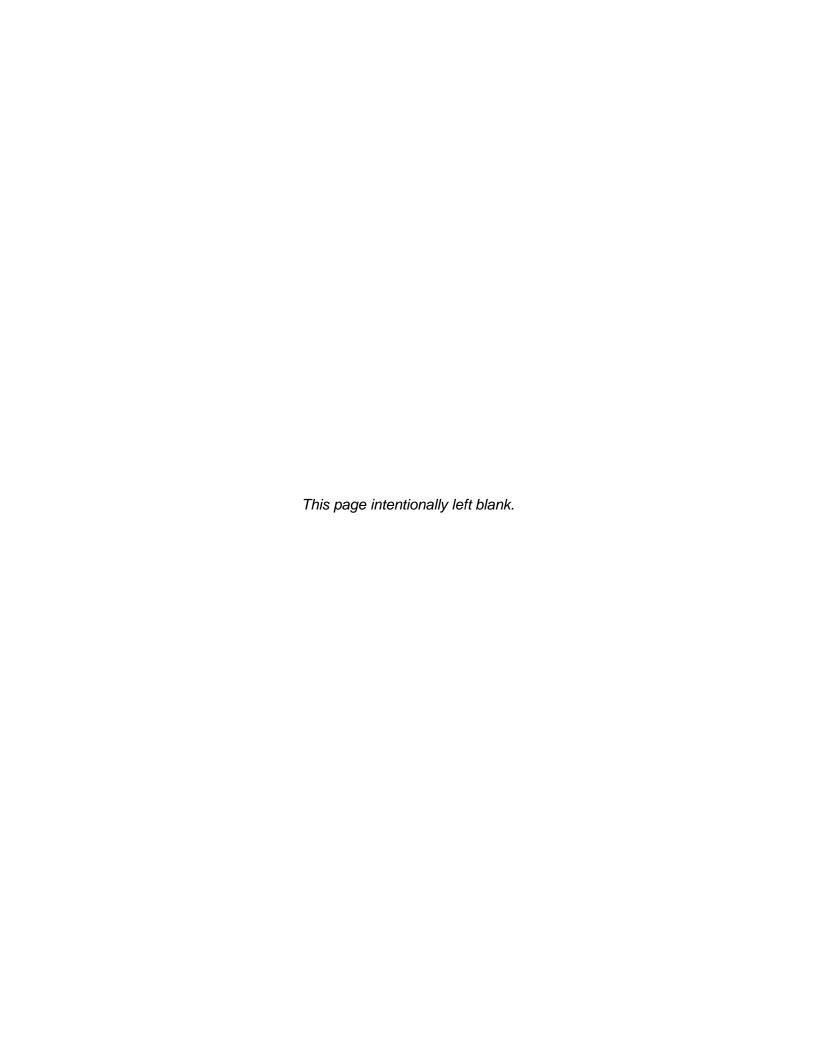
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New Tools for Echolocation Click Analysis



Appendix A: New Tools for Echolocation Click Analysis

New tools, techniques and signal classifiers have been developed recently by Bio-Waves, Inc. researchers for implementation in the widely used bioacoustic analysis software package, PAMGuard (Gillespie et al. 2008). These tools have greatly increased PAMGuard's utility and effectiveness for processing and data analysis of marine mammal tonal calls and echolocation clicks. PAMGuard is an acoustic data-processing software platform that has been widely adopted by the marine mammal bioacoustic research and mitigation & monitoring communities. PAMGuard is freely available (www.pamguard.org) and users who are familiar with the Java programming language can create custom modules to meet their needs. We work closely with the developers of PAMGuard (Sea Mammal Research Unit/University of St. Andrews) to integrate our tools and algorithms into their program.

PAMGuard contains an automated click-detector module that can be parameterized (i.e., configured) to detect clicks from specific species or species-groups. Bio-Waves, Inc. has parameterized generalized automated classifiers for sperm whales, several species of beaked whale, dolphins, and blackfish species groups. These classifiers have been tested and validated in the field during various U.S. Navy- and NOAA-funded research projects and marine mammal surveys (e.g., GOALS II [2013 Gulf of Alaska Line transect Survey], PODS [Pacific Ocean Killer Whale and Other Cetaceans Distribution Surveys], and AMAPPS [Atlantic Marine Assessment Program for Protected Species]). The classifiers also have been used for a variety of research and monitoring projects, which required efficient post-processing and analysis of large datasets from autonomous acoustic recorders. These generalized classifiers have proven reliable for both autonomous-acoustic-recorder and towed-hydrophone-array data.

In order to train classifiers to classify calls and clicks to species (within each species-group) Bio-Waves, Inc. has created a 'software bridge' between PAMGuard's click detector module and the classification module, ROCCA (Real-time Odontocete Call Classification Module). This bridge allows the click detector to pass detected clicks to ROCCA in real-time via one of two user-selected methods: 1) *Automated*: all clicks from user-selected PAMGuard species groups are sent to ROCCA or 2) *Semi-automated*: only specific clicks selected by the user are sent to ROCCA (**Figure A-1**).

Once clicks have been sent to ROCCA, new Java code written by Bio-Waves, Inc. automatically measures features (e.g., peak frequency, ICI, signal-to-noise ratio, etc.) from them. Click measurement capabilities are also available in PAMGuard Viewer Mode. 'Viewer Mode' allows efficient visual review of click detections from large datasets, by allowing data analysts to: rapidly review automated detections; select click train events; verify species identifications; and localize or re-localize individual animals. In this mode, the user manually selects click train 'events' (e.g., individual whale trains) and marks them by drawing a box around the clicks to signify an event. All of the marked clicks in the 'event' are subsequently sent to ROCCA to be measured with values saved in a database (**Figure A-2**).

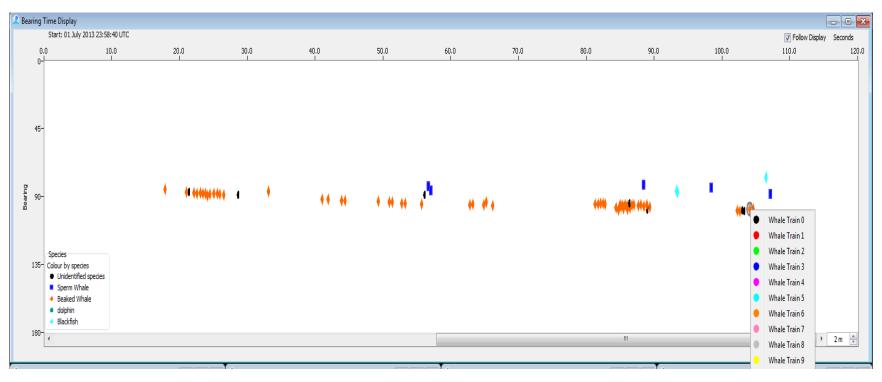


Figure A-1. PAMGuard click detector display showing the bearing (y-axis) versus time (x-axis) display with detected clicks represented as filled shapes with the color indicating automatic classification of species or species groups. Using the 'semi-automated method,' selected clicks can be manually assigned by the user to a 'whale train,' which is then sent to ROCCA for measurement. In contrast, in the 'automated method' all clicks colored as the species of interest (e.g., beaked whale (orange) would be sent to ROCCA for measurement).

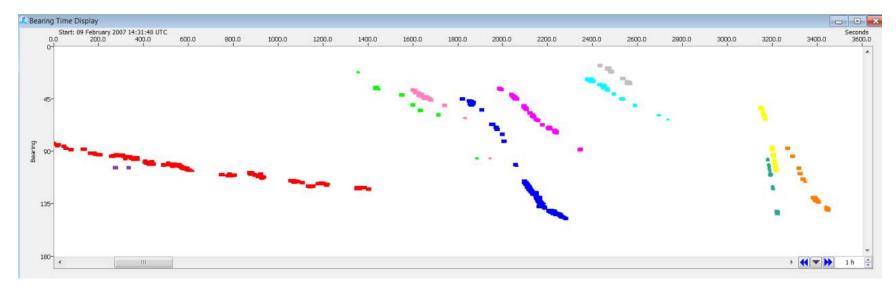


Figure A-2. PAMGuard Viewer Mode click detector display illustrating the post-processing method of sending clicks to ROCCA. 'Events' are marked as individual colors, and clicks from each event are sent to ROCCA for measurement.

Bio-Waves, Inc. researchers and programmers have also developed code in R software to automatically and efficiently provide summary statistics for measured clicks and to provide useful graphical outputs, such as histograms and boxplots (**Figures A-3** and **A-4**). This provides a way to efficiently and rapidly review and interpret results of click and call measurement, which can consist of extremely large datasets. These tools not only provide useful information about characteristics of calls and clicks but additionally, can provide analysts with the essential information for refining species group classifications to species (e.g., for beaked whales, the blackfish group [e.g., killer whales], and other species for which species--specific click measurements are well-known).

Using these methods, large data sets can be processed and analyzed quickly and effectively. For example, 1 month of continuous data can be processed in PAMGuard, with all true positive events marked by the analyst and measured in approximately 40 to 60 hours. Bio-Waves, Inc. has used these methods effectively for ground-truthing other automated detectors, reporting occurrence and distribution of beaked whales and sperm whales, and for obtaining data needed for acoustic-based density estimates of beaked whales and sperm whales.

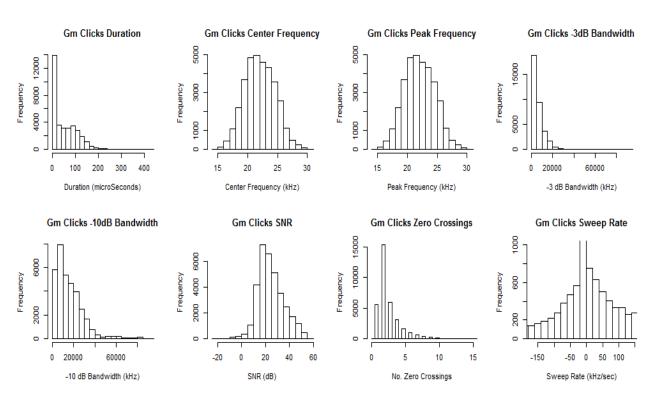


Figure A-3. Histograms showing click measurements for short-finned pilot whale (*Globicephala macrorhynchus*) clicks measured using ROCCA's click measurement tool.

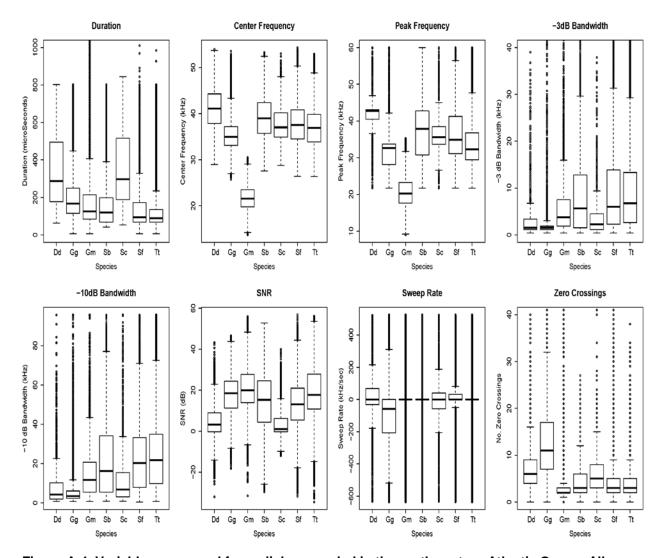


Figure A-4. Variables measured from clicks recorded in the northwestern Atlantic Ocean. All variables are shown by species as box plots. The boxes represent the upper 75 percent quartile and the lower 25 percent quartile, with the solid black horizontal line indicating the median. The hinges show the maximum and minimum values and the stars represent outliers that are more than or less than 1.5 times the quartile ranges, respectively. Species listed on the x-axis are: Delphinus delphis (Dd), Grampus griseus (Gg), Globiecephala macrorhynchus (Gm), Steno bredanensis (Sb), Stenella coeruleoalba (Sc), Stenella frontalis (Sf), and Tursiops truncatus (Tt).

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