Sound Localization Abilities of the West Indian Manatee

Final Report: January 2007

State of Florida Fish and Wildlife Conservation Commission
Project FWC 03/04-28

David Mann, University of South Florida
Gordon Bauer, New College of Florida
Debborah Colbert, Mote Marine Laboratory
Joseph Gaspard, Mote Marine Laboratory
Roger Reep, University of Florida

Permit. Research was conducted under USFWS Permit: MA837923-6.

Project Summary

The purpose of this project was to determine the ability of two captive manatees (Hugh and Buffett) to localize sound in the azimuthal plane. The project was divided into two phases. In the first phase the ability of the manatees to localize sound from four speakers arranged over 180 degrees in front from the left to the right of the subject, was measured. The purpose of this phase was to first train the manatees to discriminate among a small speaker set in a shallow water environment and to test their ability to localize a series of broad-band and tonal signals. The results of these experiments showed that performance accuracy ranged from 79-93% for Buffett, and 51-93% for Hugh for a 0.2-20 kHz broad-band signal (ranging from 200 – 3,000 ms). Three additional broad-band frequency ranges were tested (0.2 – 20, 6 – 20, and 0.2 – 2 kHz) at four durations (3,000, 1000, 500, and 200 ms). Buffett averaged 90%, 86%, and 89%, respectively on these frequency ranges over all sound durations. Hugh averaged 76%, 68%, and 65%, respectively. Both animals performed above chance levels (25% correct) with the tonal signals, which were only tested at 3,000 ms, but at a much lower accuracy than the broad-band signals. For the 4 kHz and 16 kHz tones, Hugh's accuracy was 49% and 32%, respectively, and Buffett's was 44% and 33%.

The second phase of the project measured the ability of the manatees to localize sound over 360 degrees using an 8-speaker array in the deeper end of the tank. This involved training the manatees to station midwater, and positioning the speakers farther from the manatee (3.05 m) compared to the four speaker trials (1.05 m). Sound durations were 3,000 ms for all conditions tested. The sounds were normalized to follow the published manatee audiogram with a lower sound level at higher frequencies. These experiments showed that manatees were capable of accurately localizing broad-band sounds (200-24,000 Hz) over 360 degrees The manatees performed best localizing sounds that were to the left, right or in front (Hugh: 50-100%; Buffett: 56-100% for sounds +/- 90 degrees). They performed worst on sounds located behind them, with some front-back confusions (at 180 degrees Hugh: 11%; Buffett: 63% correct). An error analysis showed that the manatees tended to make errors most commonly to the speaker to the left of the target speaker. Buffett maintained a similar level of performance over a 30 dB range (95-125 dB re 1µPa rms), however Hugh's performance fell over this range.

The ability of the two manatees to localize sounds was remarkable, given that sound travels at 1,500 m/s underwater, five times faster than in air. This reduces the inter-aural time delay that is the most important cue for sound localization in mammals. These results suggest that manatees should be able to localize the directions of oncoming boats. Continued testing will be used to determine the relative importance of low and high-frequency cues in localization.

Introduction

West Indian manatees (*Trichechus manatus*) live in turbid saltwater habitats in the summer when these waters are warm and graze primarily on sea grass (Reynolds and Odell, 1991). In the colder months, they migrate to warmer water habitats, such as freshwater springs and power plant discharge sites (Reynolds and Wilcox, 1986) where they feed primarily on water hyacinth, hydrilla, and other freshwater vegetation. The Florida manatee has been referred to as a "semi-social" species (Reynolds, 1979). They are often found grazing or traveling alone, although females with calves can be found together and large numbers of males are found with an estrous female. Manatees are threatened by naturally occurring events like cold stress and red tide, as well as human-influenced events such as boat strikes, canal lock compression, and habitat degradation (Odell and Reynolds, 1979).

The Florida manatee is protected by both the Marine Mammal Protection Act (1972) and the Endangered Species Act (1973). The January 2005 synoptic survey estimated the endangered Florida manatee population to be 3,142 (Florida Fish and Wildlife Research Institute, 2005). For many years, scientists have studied manatee ecology and population biology through field research (Hartman, 1979; U.S. Fish and Wildlife Service, 2001). As a result, numerous conservation efforts, all primarily focused on human behavior, have been initiated to help preserve this species including the installation of boater slow speed zones and manatee preservation areas. Although field research provides crucial information about the manatee's social structure, habitat usage, and migratory patterns, laboratory research is important for carefully quantifying sensory abilities.

Manatee Sensory Systems

Historically, few studies have addressed manatee sensory processes, and those that have, have tended to focus on the post-mortem physiology of the visual, tactile, and auditory systems. More recently, behavioral studies conducted with captive manatees have provided greater insight into these three sensory systems. This section briefly reviews these studies to place the underwater localization study in context of other manatee sensory systems.

Anatomical and Physiological Studies

The physiology of the manatee visual system has been investigated through the investigation of manatee eyes and visual cortex of the manatee brain. Several studies have shown that the small manatee eye has relatively few retinal ganglion cells and lacks an accommodation mechanism which suggests that the eye has most likely adapted to dim light conditions and is built for sensitivity rather than acuity (Walls, 1963 Piggins et al., 1983; West et al., 1991). Ganglion cell density suggests that the Florida manatee eye has limited resolving capability, with a minimum angle of resolution (MAR) of 20 minutes of visual arc (Mass et al., 1997). In addition, two different types of cones have been identified which suggest that manatees possess color vision (Cohen et al., 1982; Ahnelt & Bauer, unpublished data, reported in Ahnelt & Kolb, 2000).

The distribution and physiology of the manatee's vibrissae and body hairs have been investigated through post-mortem dissection. Six fields of perioral bristles, or vibrissae, have been identified on the face of the Florida manatee. Each follicle is composed of a dense connective tissue capsule with a prominent blood sinus complex and substantial innervation, which suggests that the perioral bristles play a tactile sensory role much like that of vibrissae in other mammals (Reep et al., 2001). The manatee's postcranial body hairs were also examined and all were found to contain a blood sinus and were innervated by 20-50 axons. These results suggest the possibility that manatees have a tactile system that can sense directionality in water currents, similar to that of the lateral line of fish (Reep et al., 2002).

The physiology of the manatee ear has also been studied post-mortem (Klishen et al., 1990). Heffner and Masterson (1990) studied the relationship between interaural time distances (IATD), the amount of time it takes sound to travel from one ear to the other, and upper frequency hearing limits for numerous mammals. This study found that animals with narrower heads had smaller interaural and intermeatal distances and needed higher frequency sensitivity to localize sounds. Studies of the manatee's inner ear indicate that their IATD falls between a minimum of 58 μsec when measured from intercochlear distances and a maximum of 258 μsec when measured from the external intermeatal path. This IATD is much shorter than other mammals suggesting that manatees would not have good directional hearing (Ketten et al., 1992). A more recent investigation of the zygomatic process of the squamosal bone found a lipid-filled channel, similar to that observed in the lower jaw of cetaceans, which may facilitate directional hearing (Ames et al., 2002). However, studies of evoked potentials in response to sound presentation at different body locations did not support this hypothesis (D. Mann, pers. comm.). Manatees also have a unique cochlea in which the round window is located medially, rather than laterally adjacent to the oval window as found in other mammals (B. Rommel, pers. comm). The significance of this anatomical difference is not clear.

Behavioral Studies

Vision has been investigated in three domains: brightness discrimination, color discrimination, and visual acuity. Brightness discrimination was tested with two West Indian manatees using the relative reflection of targets in a two-choice discrimination procedure. Subjects demonstrated good brightness sensitivity (Weber fraction of 0.35) similar to that of fur seals although considerably less than that of humans (Griebel & Schmid, 1997). Tests of color discrimination with West Indian manatees using a two-choice discrimination task found that manatees were able to discriminate blue and green from a series of comparably bright grays (Griebel and Schmid, 1996). Visual acuity of two Florida manatees was tested using two-choice discrimination of grating stimuli (Bauer et al., 2003). One of the subjects had an MAR of 21 minutes. Since this acuity closely agreed with the physiological estimate by Mass et al. (1997), it was considered to be species typical. The second subject's MAR was over a degree and probably represented an outlier. Physiological and behavioral measures suggest that manatees have poor visual acuity and that vision may be used as an orientation cue for large stimuli, but is not of great utility in evaluating fine details (Bauer et al., 2003).

In contrast to vision, touch appears to be remarkably acute in manatees. Observation of feeding behavior showed that the large perioral bristles located on the manatee's upper lip are used in a prehensile manner during feeding (Marshall et al., 1998 & 2003). Bachteler & Dehnhardt used a two-alternative, forced-choice paradigm to reveal excellent tactile discrimination of grating stimuli by an Antillean manatee (Weber fraction of 0.14) (Bachteler & Dehnhardt, 1999). A similar study conducted with two Florida manatees found a Weber fraction of 0.025 for one subject and 0.075 for the other (Bauer et al., 2005). These results suggest that manatee's tactile sensitivity probably plays an important sensory role for the species.

The manatee's hearing ability has been investigated using several approaches: evoked potential techniques, behavioral testing, and field testing. Early evoked-potential studies demonstrated that the largest evoked potentials occurred in the range of 1 – 1.5 kHz (Bullock et al., 1980, 1982). A later study reported a considerably higher frequency peak at 35 kHz (Popov & Supin, 1990). More recent evoked potential techniques indicated that the frequency range of detection was from 0.2 - 40 kHz (Mann et al., 2005). The temporal resolution of the manatee's auditory system was also indirectly measured using an Envelope Following Response (EFR). Results suggested that manatees could map amplitude modulated rates up 0.6 kHz, about 10 times that of humans and half that of dolphins (Mann et al., 2005).

A manatee audiogram was obtained from two Florida manatees using a forced-choice, two alternative testing paradigm (Gerstein et al., 1999). Hearing thresholds ranged from 0.5 – 38 kHz for one subject and 0.4 – 46 kHz for the other. The frequency range of best hearing for both subjects was reported to be between 6 – 20 kHz.

Manatee responses to controlled boater approaches indicated that manatees oriented towards deeper channel waters and increased their swimming speed when boats were within approximately 25 – 50 m (Nowacek et al., 2004). Although we are gaining a better understanding of the manatee's auditory detection capabilities, little work has been done on sound localization.

Sound Localization

Sound localization is the auditory system's ability to process the frequency, level, and phase of a sound and associate it with the spatial location of that sound's source (Yost, 2000). There are three dimensions from which a sound can be localized, the vertical plane also called the up-down dimension, distance also called the near-far dimension, and the horizontal plane also called the azimuth or left-right dimension. There are three cues that can be used to evaluate and process sound localization within these dimensions, interaural time, intensity, and phase differences. Interaural time differences, also known as time of arrival cues, derive from a comparison of sound arrival time at each ear. Because the speed of sound is relatively independent of frequency, variations in frequency do not have an effect on the perception of interaural time differences. Interaural intensity level differences provide cues for localization because the sound at the closest ear to a source is louder than at the further ear, due the shadowing effect of the head. The intensity level difference is dependent on the wavelength. Higher frequencies have shorter wavelengths and result in a larger sound shadow between the two ears. Interaural phase differences are interpreted when the sound that arrives at the first ear is in one period of the frequency but is out of phase when it hits the second ear. The phase difference is also dependent on the wavelength.

Terrestrial mammals differentially utilize interaural cues. The hedgehog (Masterson et al., 1975) and the Northern grasshopper mouse (Heffner & Heffner, 1988) both seem to have reduced or lost the ability to utilize phase difference cues. In addition, the elephant, horse, pig, goat and cattle (Heffner & Heffner, 1982, 1984, 1989, and 1992b) appear to have reduced or lost the ability to use interaural intensity level difference cues. At least one species, the pocket gopher (*Geomys bursarius*), is known to be incapable of using any of the interaural time, intensity, and phase difference cues (Heffner & Heffner, 1990). Burda (1990) suggested that this may be a result of this fossorial species' adaptation to living in an underground environment where azimuth cues have little meaning. Although similar primitive species have not been studied, they may also lack the use of interaural differential cues. While in-air localization may be difficult or impossible for some terrestrial species, the ability to localize sounds underwater may present even more of a challenge to marine mammals because of the greater speed of sound underwater.

In-Air vs. In-Water Acoustic Properties

The speed of sound in water (1500 m/sec) is almost five times faster than in air (340 m/sec) (Urick, 1996). Therefore, marine mammal auditory systems need to be able to process the frequency, intensity, and phase of sounds that move almost five times faster than the sounds which terrestrial mammals need to process. Acoustic energy in water propagates more efficiently than light, thermal or electromagnetic energy, which attenuate rapidly, therefore underwater sound localization may provide the best means for marine mammals to find food and conspecifics while avoiding predation in deep water (Au, 1993). Shallow water conditions, however, present additional acoustical challenges. Acoustic energy in shallow water does not travel as efficiently as it does in deep water with a lot of acoustic multipaths (Medwin and Clay, 1998). Since Florida manatees spend a significant amount of time grazing in shallow water, localization of sound from conspecifics and boats may be particularly challenging.

Behavioral tests of underwater sound localization abilities have been conducted with numerous species of captive marine mammals including sea lions (Gentry, 1967; Moore, 1974; Moore & Au, 1975; Holt et al., 2004), bottlenose dolphins (Renaud & Popper, 1975; Moore & Pawloski, 1993; Moore and Brill, 2001), and harbor seals (Anderson, 1970; Terhune, 1974). These studies suggest that marine mammals have the ability to localize underwater sounds by using the same interaural, differential sound cues that terrestrial mammals use.

To date, only one behavioral study has been conducted to measure a Florida manatee's ability to localize specific sounds (Gerstein, 1999). The subject localized a 20 ms tonal signal of 0.5, 1.6, 3, 6, or 12 kHz, which was pulsed for durations of either 200 or 500 ms, paused for 400 ms, and then repeated, from one of four locations. Two speakers were positioned at 45^{o} and two at 90^{o} angles to the manatee's head. Results indicated that the manatee was able to localize all of the signals, but that accuracy decreased with the lower frequencies. Accuracy was better with the longer durations and at the 45^{o} angles. Based upon these results, Gerstein suggested that manatees may not be able to effectively localize the low frequency sounds of boat engines to avoid collisions in the wild.

Florida Manatees and Environmental Noise

The Florida manatee lives in an environment where recreational boats are found in high numbers and conspecifics are often out of visual range. How then do they avoid boat collisions and find conspecifics? Manatee visual acuity appears to be poorly adapted for these tasks, and although their tactile sensitivity is impressive, it is probably best utilized to investigate objects nearby. Neuroanatomical data suggest their chemical senses are of secondary importance. Therefore, the manatee auditory system may play a critical role in accomplishing these challenging tasks.

The Florida manatee shares its habitat with over 1,000,000 registered boats (Florida Department of Highway Safety and Motor Vehicles). In 2004, watercraft-related injuries accounted for 25% of all manatee mortalities, (Florida Fish and Wildlife Research Institute, 2004). One question that has been raised is, if manatees can localize boat engine noise, why are there so many watercraft-related mortalities? Conversely, if manatees cannot localize boat engine noise, why are there not more watercraft-related mortalities? Boat engine noise can be categorized in two ways: cavitating, which is associated with propeller rotation and produces higher frequency broad-band noise, and non-cavitating, which is associated with other propulsion machinery that produces lower frequency broad-band noise. Boats traveling at high speeds usually produce the higher frequency cavitating noise, while boats traveling at idle and slow speeds produce the lower frequency non-cavitating noise (Ross, 1976). The dominant recreational boat frequency range is typically 0.01 – 2 kHz but can reach over 20 kHz. The estimated 1/3-octave source levels at 1 m for small motorboats are 120-160 dB re 1 µPa-m (Gerstein, 2002; Richardson et al., 1995). Personal watercraft, such as jet-skis, utilize jet propulsion rather than outboard propellers and are approximately 9 dB quieter than small motorboats (Buckstaff, 2004).

The often solitary Florida manatee is able to find conspecifics in a wide-ranging habitat. The question of how these semi-social animals find one another remains unanswered, but the auditory localization of vocalizations provides a good candidate solution. Vocalizations for both subspecies of West Indian manatee (Florida and Antillean) are short duration, tonal harmonic complexes that range from almost pure tones to broad-band noise and have a fundamental frequency that ranges from 2.5 – 5.9 kHz but can extend to 15 kHz (Nowacek, et al., 2003).

The question of whether or not manatees are able to localize boat engine noises and conspecific vocalizations has puzzled researchers for years and is a topic of debate that warrants further investigation. In this study, two Florida manatees were tested to determine their ability to localize sounds of different frequencies and durations in the azimuthal plane.

Methods

Subjects

The subjects were two captive-born male Florida manatees (*Trichechus manatus latirostris*) that reside at Mote Marine Laboratory and Aquarium in Sarasota, Florida. All procedures used were permitted through the United States Fish and Wildlife Service (Permit # MA837923-6) and approved by the Institutional Animal Care and Use Committee of Mote Marine Laboratory and Aquarium. At the inception of this study Hugh was 20 years of age, weighed 547 kg, and was 304 cm in length, while Buffett was 17 years of age, weighed 773 kg, and was 334 cm in length. They were housed in a 265,000 liter exhibit that was composed of three inter-connected sections: a 3.6 x 4.5 x 1.5 m Medical Pool, a 4.3 x 4.9 x 1.5 m Shelf Area, and a 9.1 x 9.1 x 3 m Exhibit Area (Figure 1). Both animals had acquired an extensive training history over the previous seven years and were subjects in an auditory evoked potential study (Mann et al., 2005), making them excellent candidates for this project. In addition, they had been trained for husbandry procedures (Colbert et al., 2001) and studies of serum and urine creatinine (Manire et al., 2003), visual acuity (Bauer et al., 2003), lung capacity (Kirkpatrick et al., 2002), and tactile sensitivity (Bauer et al., 2005).

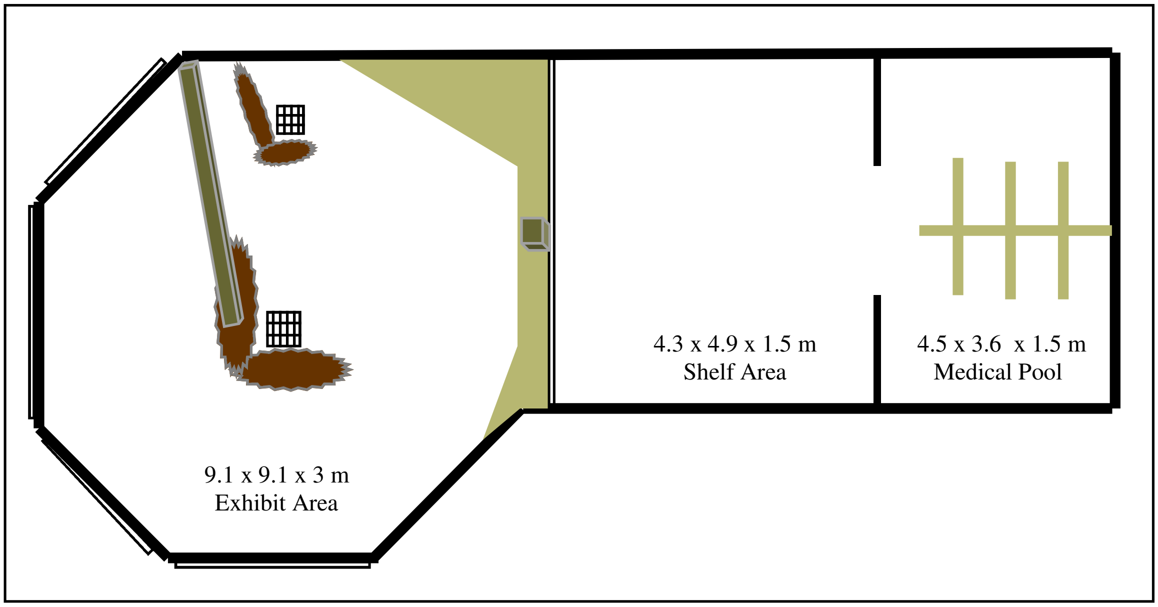

Figure 1. Diagram of the 265,000 L manatee exhibit composed of a Medical Pool, Shelf Area, and Exhibit Area. The lines in the Medical Pool represent a distance scale, used in a previous study that was painted on the floor of the exhibit. The oval masses in the Exhibit Areas represent outcroppings in the bottom terrain (built of cement) to conceal the two floor-level filtration drains (gratings). The rectangles represent a tree log and stump (built of cement).

Overview

This section provides a synopsis of the two protocols used in testing, a 4-speaker array and an 8-speaker array.

Experimental Design

4-speaker array

Testing for the 4-speaker array was conducted in the center of the Shelf Area (Figure 1) with the test animal positioned midway between the exhibit bottom and the surface of the water (approximately .75 m below the surface). The non-test animal was held at station in either the Medical Pool or the Deep Area. Four test speakers were positioned to the front 180 degrees at 45, 90, 270, and 315 degrees relative to the front of the subject's head at a distance of 105 cm (Figure 2). The test subject was required to position himself perpendicular to a stationing bar with the crease of his rostrum, approximately 10 cm posterior to his nostrils, pressed against it, in response to a specific pulsed-tone played from a stationing speaker. The manatee remained stationed until a test signal was played from one of four underwater speakers. He swam to and depressed the speaker from which the sound originated. If correct, a secondary reinforcer tone was emitted from the test speaker and the subject returned to the stationing device to be fed a primary reinforcement of food. If incorrect, the stationing tone was played from the stationing apparatus speaker and the subject re-positioned correctly with no primary or secondary reinforcement given, and awaited the initiation of the next trial. All test trials were video-recorded from an overhead camera.

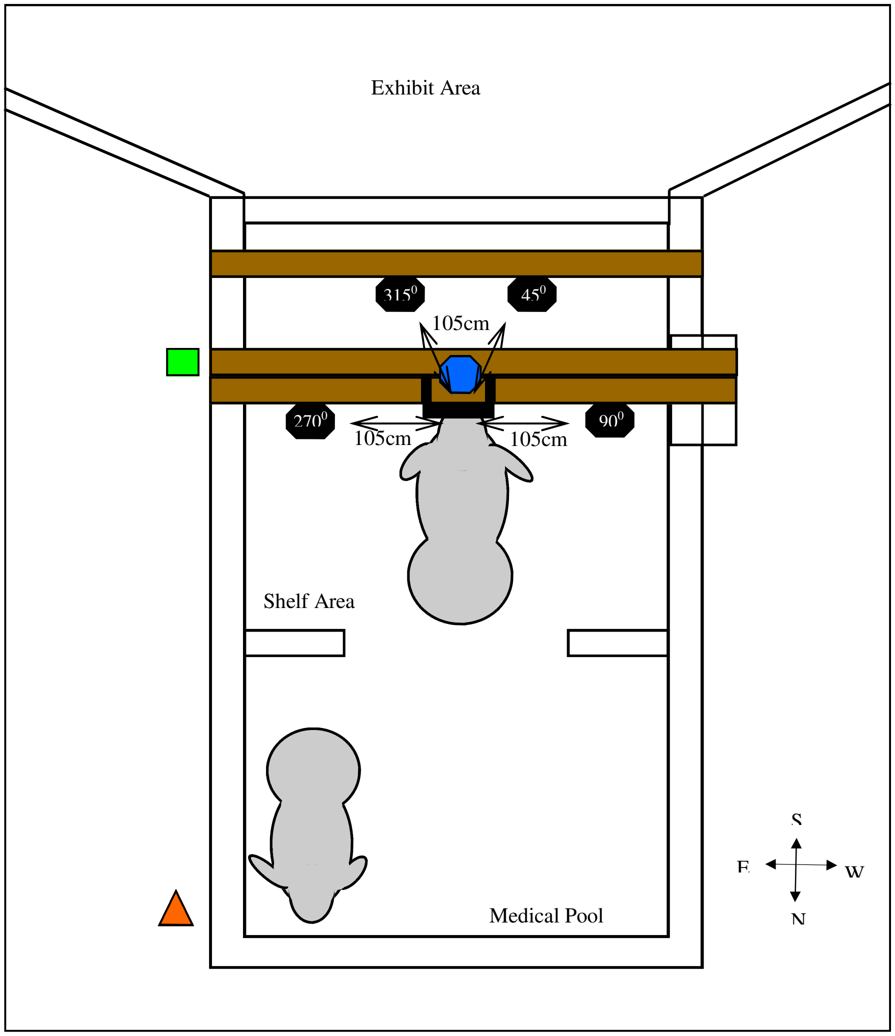

Figure 2. Testing set up for the four-speaker array.

8-Speaker Array

Testing for the 8-speaker array was conducted in the deeper Exhibit Area (Figure 3). A trainer was positioned on a catwalk across the exhibit area and the manatee stationed on a PVC apparatus 1.37 m below the surface of the water. Eight test speakers were positioned with equal spacing over 360 degrees at 0, 45, 90, 135, 180, 225, 270, 315 degrees relative to the front of the subject's head at a distance of 3.05 m (Figure 3). The same procedural design as the 4-speaker array testing was used for the 8-speaker array testing.

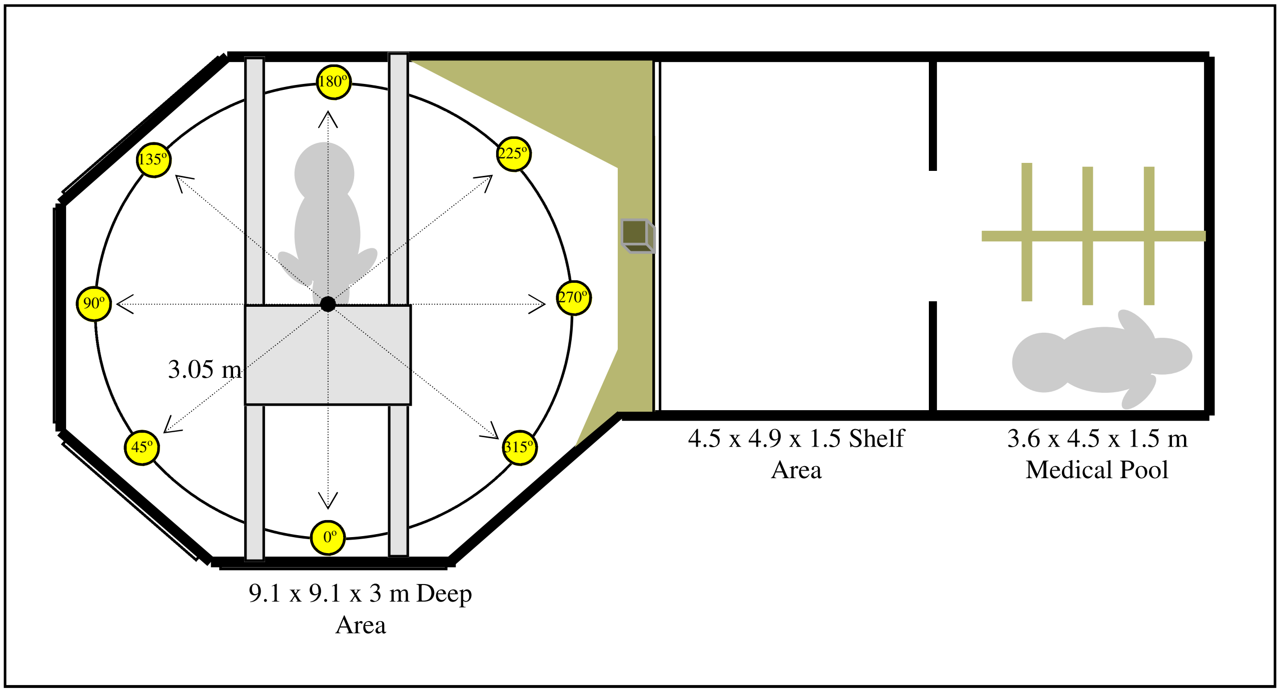

Figure 3. Testing setup for the 8-speaker array.

Signal Generation

Signals were generated digitally by a Tucker-Davis Technologies (TDT) real-time processor (RP2.1), and attenuated with a programmable attenuator (PA5) to control intensity level. The signals were amplified with a Hafler power amplifier and switched through a power multiplexer (PM2R) that was capable of switching the signal to be presented by one of the testing speakers (AquaSynthesis). The status of a switch at each speaker location was monitored by the digital inputs on two RP2's. A separate digital to analog channel was used to generate the signal to the stationing speaker at the center of the array.

Computer and Programming

A Dell laptop computer (model Latitude D505) with Windows XP was used to run the signal generation equipment and to automatically download the parameters of each trial into an Excel file.

All signals and conditions of the testing sequence used in the experiments were programmed in the RPvds language for the TDT RP2.1. These programs included the development of two "call to station" signals (one unique to each subject), five "test/training" signals, and two "reinforcement" signals (one unique to each subject) used to bridge the subject if he was correct.

A program was also developed to generate blocks of twelve trials that were counterbalanced between the speaker locations, with a criterion of no more than two trials in a row from the same location.

Each trial was initiated and completed through an electronic button box which was connected to the RP2 unit, and then into the laptop computer (Figure 4). Four LEDs were included on the button box to provide a visual indication that their corresponding signals were played and that the trial was downloaded into an Excel file.

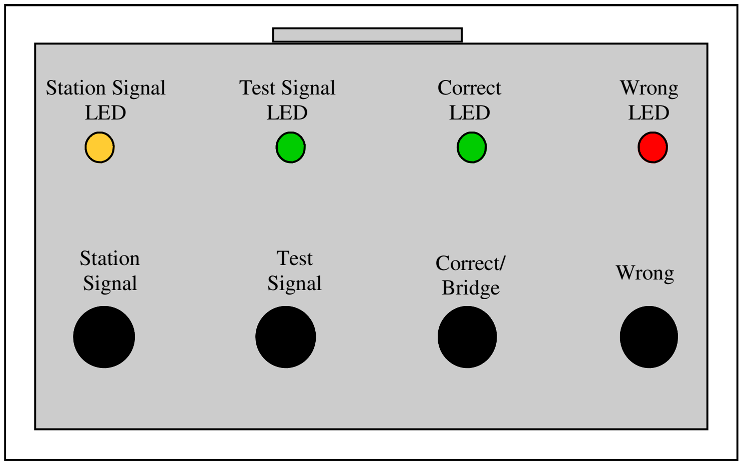

Figure 4: Electronic button box used to run the sessions and automatically download each trial into a digital Excel file.

Personnel

Three people ran the experiment; a "Test Trainer" who worked with the test subject, a "Data-recorder" who ran the computer and recorded the data, and an "Auxiliary Trainer" who worked with the non-test manatee. These individuals had extensive experience working with the subjects and were completely familiar with the computer program, the experimental plan, and training procedures.

The Data-recorder was seated behind the computer out of both the test subject's and Test Trainer's line of sight to avoid inadvertent cuing of the Test Trainer. From this location, the Data-recorder was unable to determine the test subject's position in the water.

The Test Trainer was positioned on the center of wooden boards that were suspended across the Shelf Area for the 4-speaker tests or on the catwalk above the exhibit area for the 8-speaker tests, directly above the stationing apparatus. The Test Trainer was "blind" to the test stimulus locations and wore sound-dampening headphones to avoid cueing the subject. The Auxiliary trainer was positioned at either the northeast end of the Medical Pool or Deep Area, out of the test subject's line of sight.

General Training Procedures

Training and testing sessions were run between 0700 and 1030h, five days per week before the Aquarium was open to the public. The manatees' daily ration of food (72 heads of romaine lettuce and 12 bunches of kale) was fed to the animals from 1200 to 1400 h and was usually consumed by 1700 h, leaving a 14 to 16 hour overnight fast before training was initiated the following morning. All training was completed using standard classical and operant conditioning techniques. The primary reinforcers used included bite size pieces of apples, beets and baby peeled carrots. Zupreem monkey biscuits, one of the manatees preferred foods, were used to reward an especially desired behavior during shaping procedures. A unique whistle was used as a secondary reinforcer to bridge each animal independently of one another. In addition, verbal and tactile secondary reinforcers were used. Shaping by reinforcement of successive approximations was used to train all behaviors (Pepper & Defran, 1975) and undesirable behaviors were ignored. In addition, time-outs (Pepper & Defran, 1975; Domjan, 1998, p 272), or the removal of the opportunity to receive reinforcement, were used if a string of undesirable behaviors occurred.

Initial Training: East Wall of the Shelf

Stationing

Training was initiated with the subject positioned perpendicular to the east wall of the Shelf Area and the other animal positioned in the northwest corner of the Medical Pool (Figure 5). The subject was positioned in this way to better allow the Test Trainer to easily reach the subject and physically maneuver his body and head into the correct position.

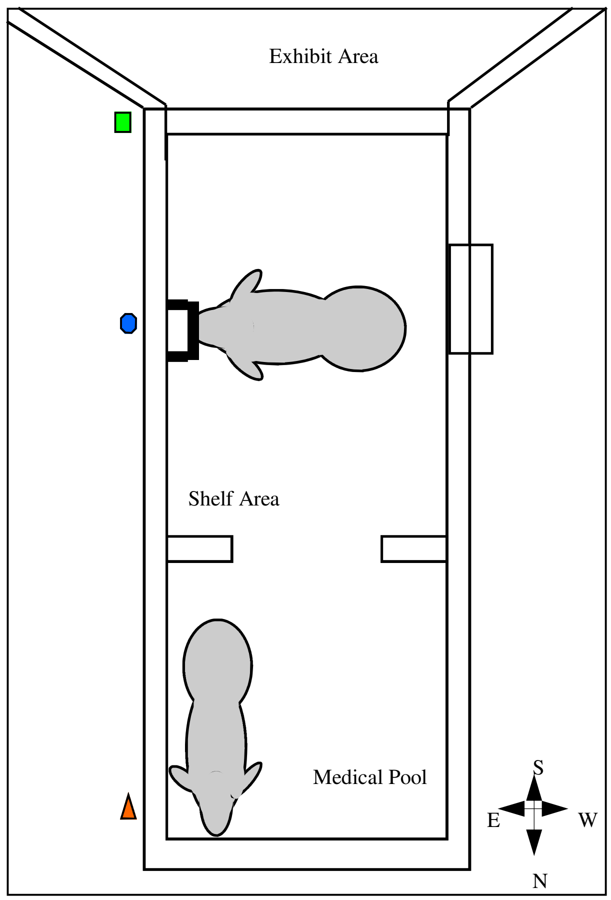

Figure 5. Training configuration for the east wall of the Shelf Area. The blue octagon represents the Test Trainer's location, the green square represents the Data-recorder's location, and the orange triangle represents the Auxiliary trainer's location.

A 2.54 cm diameter polyvinyl choloride (PVC) stationing apparatus was designed to fit over the edge of the exhibit wall for stability (Figure 6). It extended 30.48 cm below the surface of the water. To prevent interference between the sound source and the subject's ears, the stationing apparatus had a 23 cm stationing bar that the subject pushed his rostrum against instead of the commonly used hoop or frame that encircles the head completely. An underwater speaker was suspended from the top of the stationing apparatus and positioned above the manatee's head, just below the surface of the water.

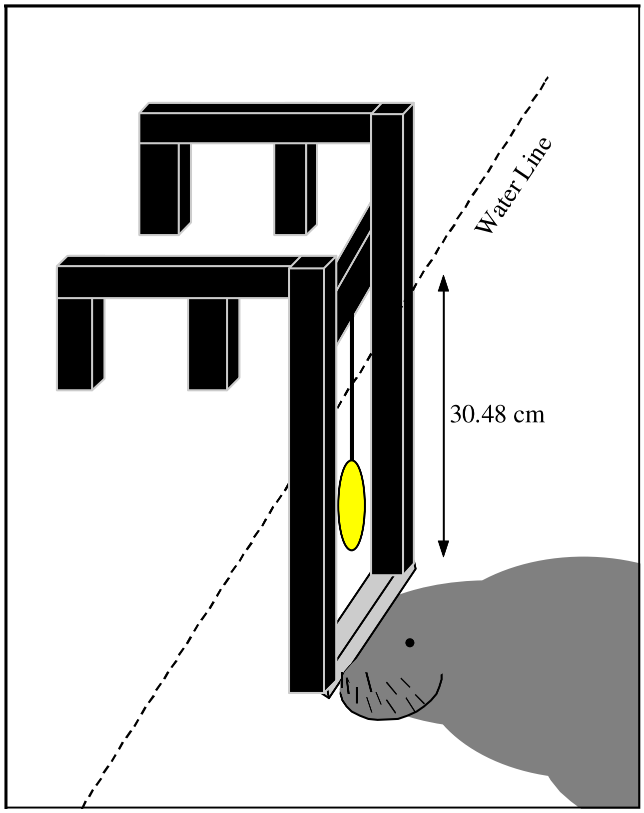

Figure 6. Stationing apparatus. The yellow circle represents the speaker that played the stationing tones. The subject pressed the crease of his rostrum up against the gray stationing bar on the bottom.

A unique sound signal called the subject to station. Each subject's station signal ranged from 10 to 20 kHz and played for a 2000 ms duration, however Buffett's repeated at a slower rate of 1.5/s while Hugh's repeated at a faster rate of 5/s. In response to their station signal, each subject was trained to swim to the stationing apparatus, position himself perpendicular to it, and press the crease of his rostrum against the stationing bar. Both animals had previously been trained to station and follow their own individual targets, and in the early stages of shaping the stationing behavior, the Test Trainer used the subject's target to guide him to the stationing bar when the station signal was played, and in some cases manual manipulation was used. Over the course of multiple sessions, each learned to station correctly in response to the signal. The length of time spent at station was extended to 60 seconds using a fixed-interval schedule of reinforcement (Ramirez, 1999).

Speaker Localization

Once the manatees had a firm grasp of the stationing behavior, training for speaker localization was initiated. The speakers were suspended from speaker holders made of 1.88 cm diameter PVC pipe. The speaker was attached to a long suspension rod which was designed to pivot so that the speaker at the bottom of the rod could be pushed backwards while the top of the rod tilted forward to touch the speaker holder frame like a pendulum (Figure 7). Initially, one test speaker was placed approximately 20 cm away from the subject's head at either a 90^{o} or 270^{o} angle (subject's head was facing 0^{o}). This speaker's position was alternated between the two sides so that the subject became familiar with moving to either the left or right to select the correct location.

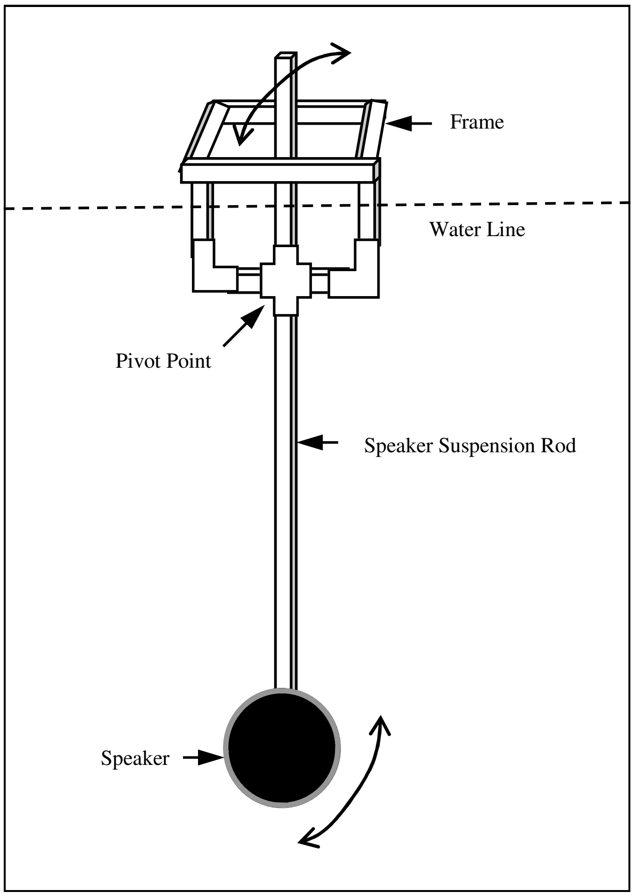

Figure 7. Speaker holder with attached underwater speaker. The speaker suspension rod was designed to pivot so that the manatee could push the speaker at the bottom backward while the top tilted forward to touch the speaker holder frame.

A 3000 ms broad-band signal with a frequency range of 0.2 – 20 kHz was programmed in RPvds and used to train this discrimination task. Two secondary reinforcement signals were programmed to match the unique whistles used to bridge each animal (Figure 8).

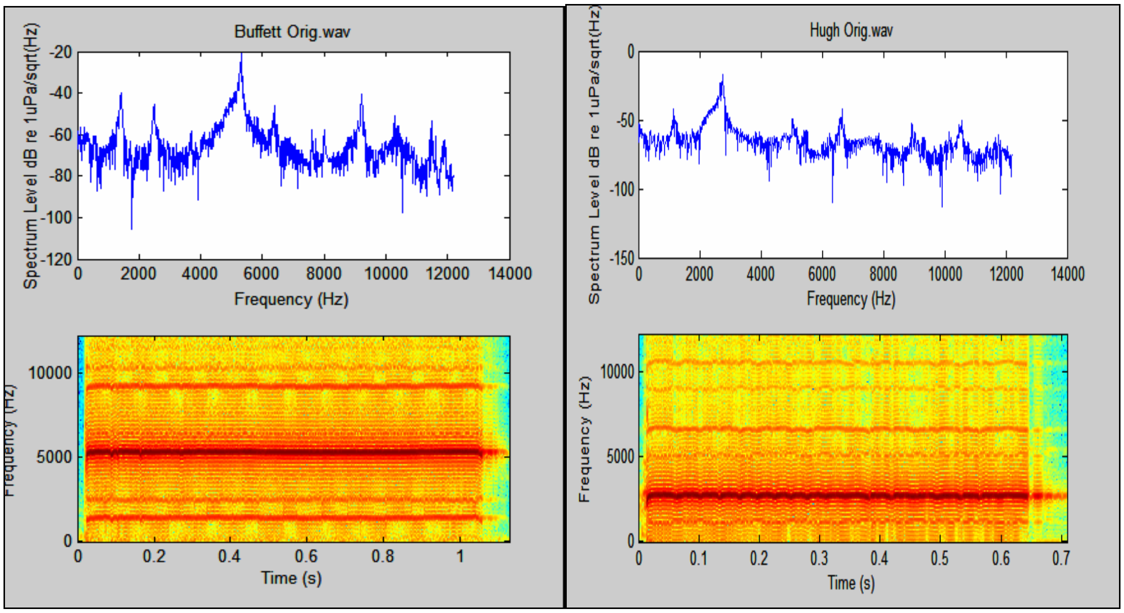

Figure 8. Power spectra (top) and spectrograms (bottom) of the secondary reinforcement signals. Buffett's had a peak at 5.3 kHz, while Hugh's had more of a warble to it with a peak at 2.7 kHz.

When the subject had stationed correctly, the training signal was played and the Test Trainer used the subject's target or manual guidance to direct the subject to the test speaker. When the subject touched the test speaker with his rostrum, his secondary reinforcer signal was played from the test speaker and he was rewarded with food. Initially, the training signal was played continuously until the subject pressed the test speaker. Once the desired behavior was obtained, the sound duration was shortened to a series of 1000 ms signals, and finally reduced to a single 1000 ms signal. Through successive approximations, the subjects were trained to approach and push the test speaker backwards until the top of the speaker suspension rod touched the front of the speaker holder frame when the training signal was played. In addition, the distance of the speaker from the manatee's head was gradually increased from 20 cm to 105 cm.

After each subject learned to select the single test speaker when it was located 100 cm away and was re-positioned at either the 90^{o} or 270^{o} location in relation to the manatee's orientation (0^{o}), a second test speaker was introduced so that two test speakers were always present, one at 90^{o} and one at the 270^{o} (Figure 9). The manatees were then required to localize the training signal source in a two-choice discrimination format. The training signal was delivered through one of the two test speakers based on counterbalanced schedules (Gellermann, 1933; Fellows, 1967) that were programmed in RPvds.

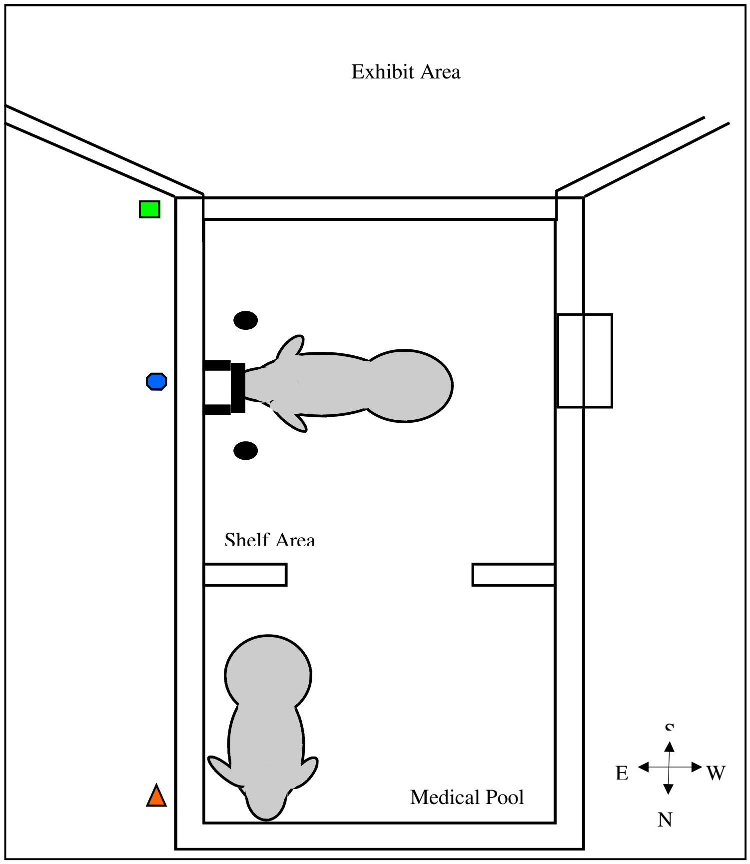

Figure 9. Training configuration for the east wall of the Shelf Area with two speaker locations. The black circles represent the speakers. The blue octagon represents the Test Trainer's location, the green square represents the Data-recorder's location, and the orange triangle represents the Auxiliary Trainer's location.

When the subjects were able to dependably perform the discrimination task at above a 75% accuracy level, the stationing apparatus and speakers were modified to reach a depth of 0.75 m, the mid-way point of the water depth in the Shelf Area, to equalize the amount of sound reflectivity from the surface and the bottom. The previously established behaviors were re-shaped to meet the new depth requirements.

Training Phase I: 4-Speaker Array

When the subjects reliably discriminated between the two test speakers located at the east wall of the Shelf Area at 0.75 m, the set-up was moved to the center of the Shelf Area and rotated 90^{o} to the South. The Test Trainer was positioned on a wooden bridge 15.24 cm above the surface of the water and spanning the width of the Shelf Area. Training was re-established in this new location and when subjects performed reliably at a 75% accuracy level or higher, a third test speaker was introduced and positioned at a 0^{o} angle to the animal's head (Figure 10).

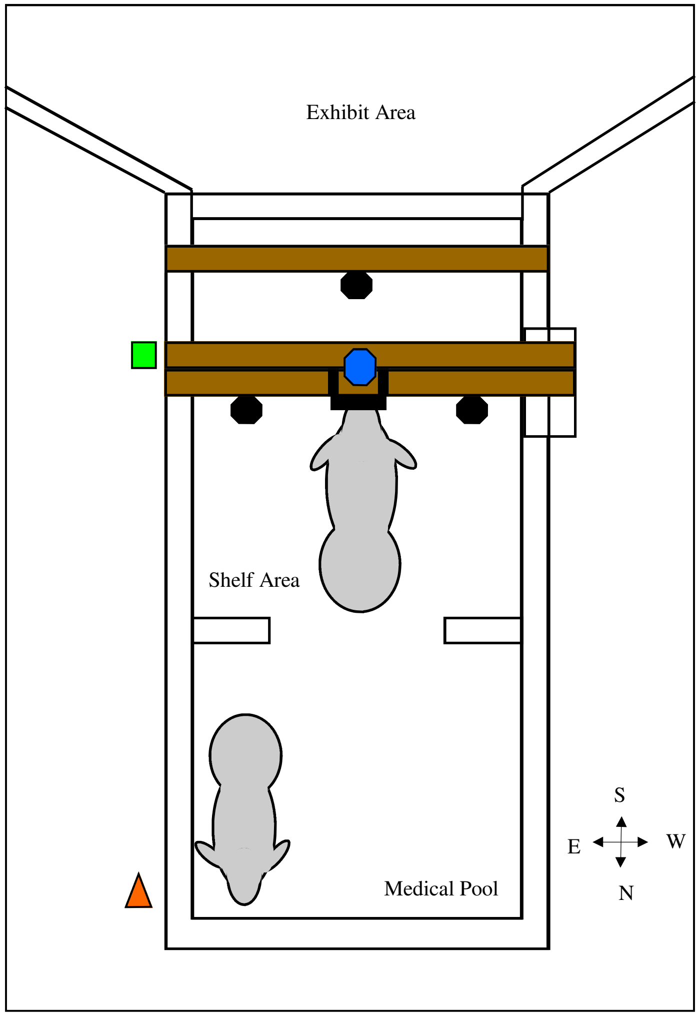

Figure 10. Training configuration for the center of the Shelf Area with three speakers represented as the black circles. The blue octagon represents the Test Trainer's location, the green square represents the Data-recorder's location, and the orange triangle represents the Auxiliary Trainer's location.

It quickly became apparent that the subjects had difficulty localizing the training signal, especially when it originated from the 0^o location. Both subjects' performance accuracy declined and they demonstrated various signs of frustration including chuffing, leaving the task multiple times, as well as grabbing, pulling, and breaking the equipment. The training signal duration was increased from 1000 ms to 3000 ms to simplify this part of the training and reduce the animals' level of frustration and their performance improved slightly but was still poor with the speaker located at 0^o. This finding was not surprising considering that even humans have difficulty localizing sound when it is directly in front or back of them because there are no interaural differences in time of arrival cues (Yost, 2000).

Therefore, the speaker directly in front of the subject was eliminated and a four speaker array was employed. The four speakers were permanently positioned 105 cm from the center of the subject's stationing bar, at 45°, 90°, 270°, and 315° angles (Figure 11). Training for the localization task continued until each subject performed the task at 75% correct or higher for a minimum of 72 consecutive trials.

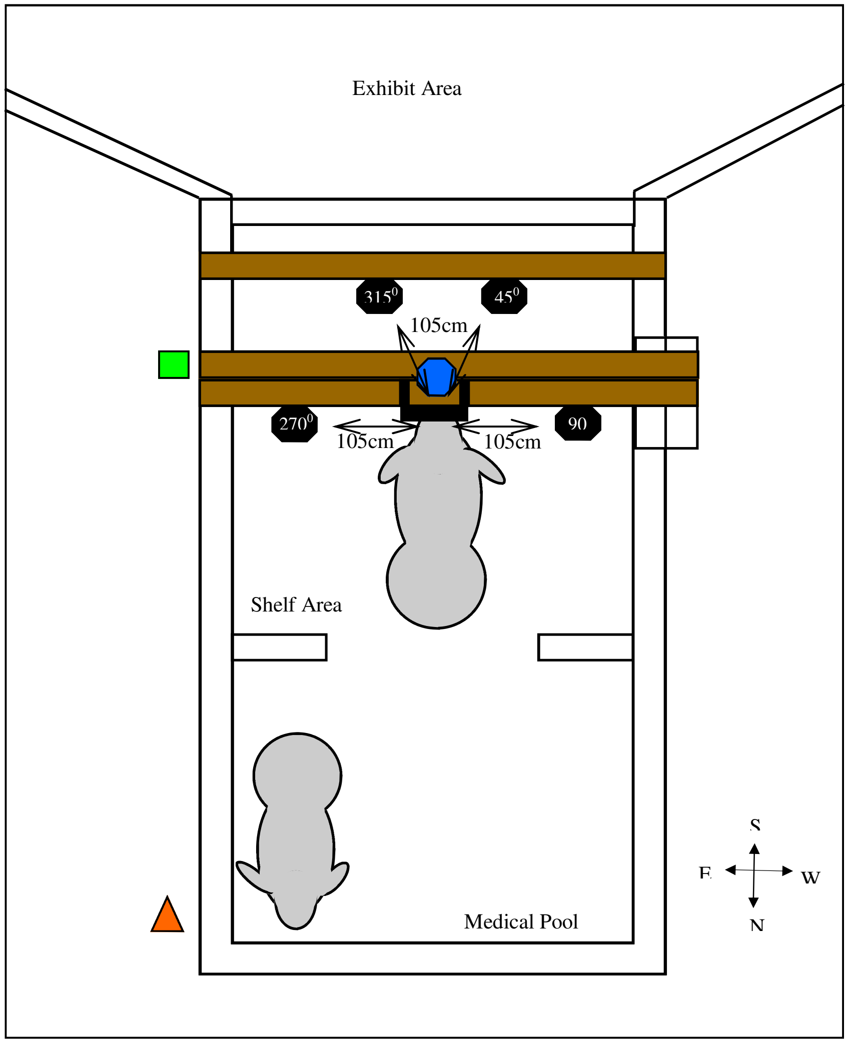

Figure 11. Training configuration for the center of the Shelf Area with four test speakers, located 105 cm from the center of the stationing bar and 0.75 m below the surface. The test speakers are represented as the black circles. The blue octagon represents the Test Trainer's location, the green square represents the Data-recorder's location, and the orange triangle represents the Auxiliary Trainer's location.

Training Phase II: 8-Speaker Array

After completion of testing with the 4-speaker array, training for the 8-speaker array was initiated. This move involved several substantive changes for the manatees:

1) They stationed at greater depth, 1.37 m, and the speakers were located at this new depth. This required greater buoyancy control than we had previously required from our

subjects and took some practice to realize. 2) The distance to the speakers increased from 1.05 m to 3.05 m. This led to a substantial increase in distance and time between the beginning of a trial, the response, and reinforcement; a substantial training problem. 3) Twice as many speakers were used. 4) Some speakers were located behind the subjects and were therefore not visible when test sounds were played (Figure 12).

A quick test for transfer from the 4-speaker to the 8-speaker array indicated that the new protocol would have to be trained in stages. The stationing response was worked on separately from touching the test speakers. When performance was stable on both, the behaviors were chained (i.e., linked). Swimming to the more distant speakers was handled initially by playing the test sounds for long periods (e.g., 10 s) and then reducing the duration in steps to 3 s. The number of speakers presented was increased in steps from 4 to 6 to 8 speakers.

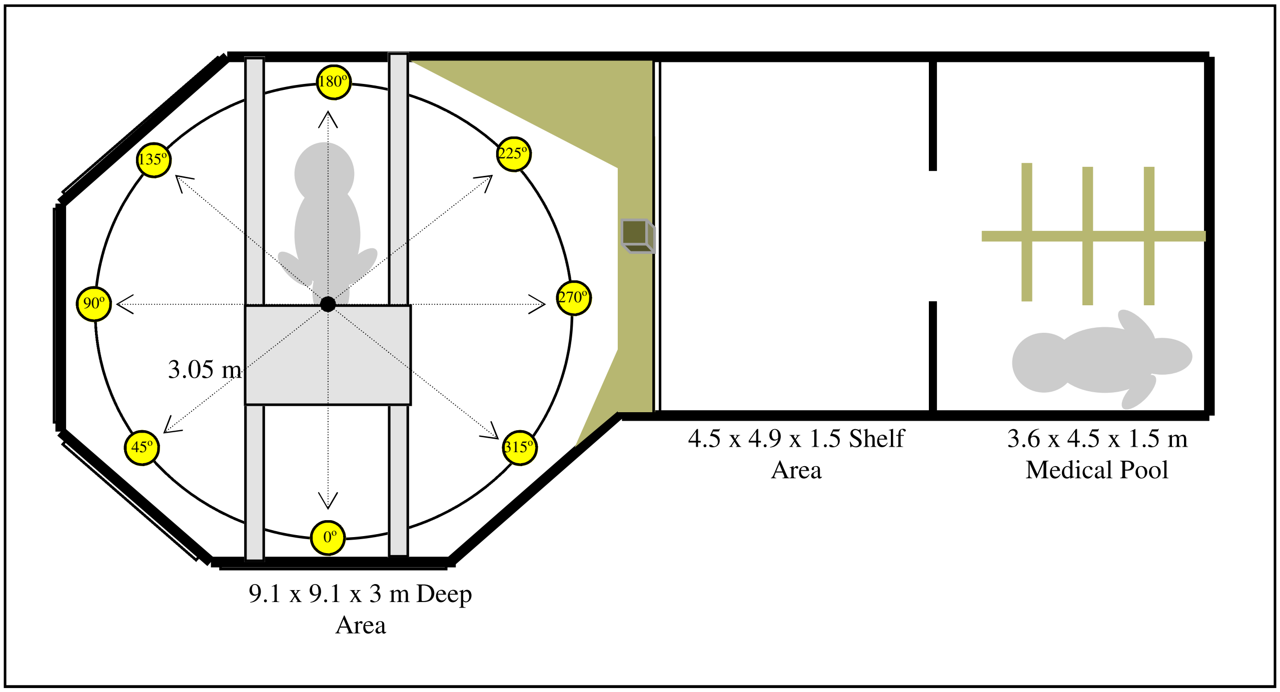

Figure 12. Eight speakers were used in the Exhibit Area. The manatee stationed in the center of the array with the speakers positioned every 45°.

Testing Procedures

This section provides specific information about the experimental conditions, sound calibrations, ambient sound conditions, experimental controls, and test block criteria. Additionally, computer programming, personnel responsibilities, and data and video recording methodology are expanded upon. To summarize procedures, a step-by-step test trial sequence is presented.

Experimental Conditions

Fourteen experimental conditions were tested for the 4-speaker experiments (Table 1). These included three broad-band noise bursts of different frequency ranges tested at four durations, and two tonal signals tested at one duration. All of the test sounds were played at the same spectrum level. Because of this, sounds with broader frequency spectra had louder root mean square (rms) amplitudes. The sound levels were also randomized ±1.5 dB to obscure any loudness differences between speakers. Six blocks of twelve, counterbalanced trials were run for each of the 14 conditions.

Table 1
Conditions tested in the four speaker trials included three broad-band signals at four durations and two tonal signals at one duration. All signals were played at the same spectrum level.

Frequency (kHz)	Duration (ms)	Spectrum Level (dB re 1 uPa)
0.2 - 20	3000, 1000, 500, 200	100
6 - 20	3000, 1000, 500, 200	100
0.2 - 2	3000, 1000, 500, 200	100
4	3000	100
16	3000	100

Broad-band sounds are more typically found in all natural habitats than tonal signals. The 0.2 - 20 kHz condition had the widest range of frequencies included in it and was the broad-band signal used during training, the 6 - 20 kHz condition contained the highest frequencies, and the 0.2 – 2 kHz condition contained the lowest frequencies.

The testing of tonal signals examined the manatees' ability to localize sounds that were frequency specific. The low frequency 4 kHz tone was similar to the dominant frequency of manatee vocalizations while the high frequency 16 kHz tone was comparable to the manatees' peak hearing frequency.

The order of test stimuli presentation was as follows: six blocks of each broad-band noise, starting with 0.2 – 20 kHz, then 6 – 20 kHz, and finally 0.2 – 2 kHz, were tested at the 3000 ms duration. This order was followed throughout each of the three other sound duration conditions (1000, 500, 200 ms). The tonal signals were only tested at the 3000 ms duration because the subjects performed at a much lower accuracy level and were exhibiting strong signs of frustration.

For the 8-speaker array a block of trials consisted of 16 counterbalanced trials with two presentations at each speaker. At this point, we report only results for one broadband stimulus, 0.2- 24 kHz, which was tested over a range of sound levels.

Personnel Responsibilities

Personnel tasks were designed to prevent possible cuing confounds, while still executing the testing efficiently. The Data-recorder, who was positioned out of sight of the Test Trainer and the subject, had six duties. The first was to set up the correct experimental conditions needed for the different portions of the session on the computer using a graphical user interface that was programmed in Visual C (see Appendix A for experimental condition set up protocols). The second was to initiate each trial through the button box when instructed to do so by the Test Trainer. The third was to determine which location the subject selected from the Test Trainer and to inform the Test Trainer if this location was correct by leaning out from behind the computer to give a head nod, or if wrong to give a head shake. The fourth was to complete the trial through the button box. The fifth was to record all data on a tank-side session sheet. The sixth was to run the video equipment.

The Test Trainer, who was blind to the test signal locations and wore sound-dampening head phones, was responsible for six duties. The first was to ensure that the subject was positioned correctly before the initiation of each trial. The second was to ensure that a 25 second minimum inter-trial interval was met. The third was to let the Data-recorder know when to initiate each trial by verbally stating “tone”. The fourth was to let the Data-recorder know which location the animal selected by verbally stating the speaker that the animal chose. The fifth was to determine if the subject was correct by looking at the Data-recorder when she came into view for the appropriate head nod or shake. The sixth was to provide the subject his primary reinforcement if he was correct and to let the Data-recorder know when the next trial could be initiated by verbally stating “station”.

The Auxiliary trainer was responsible for holding the non-test animal at station in either the northeast corner of the Medical Pool or Exhibit Area throughout the test animal’s session. The non-test animal was positioned out of view of the test animal and could be held in either a dorsal-up or ventral-up layout position.

Data Recording

Data from each session were recorded in three ways. The first was through the automated computer reports that were recorded in a digital format within the Excel file. The second was through hand-recorded reports that the Data-recorder completed on a tank-side data sheet. This information was then manually entered into a Microsoft Access database created on a Dell desktop computer (model Dimension 8300) after the completion of each session. All data entered into the database was double-checked for accuracy by a second trainer after they were entered. This database was designed specifically for this experiment and had a user-friendly data entry screen (Figure 13). This data base was then used to compile and analyze the test data. The third data recording method was through the video recording of each test block. A Sony variable zoom, high resolution, outdoor weather proof, color dome camera (model SCW-CD358DVP) was attached to the exhibit’s ceiling directly over the subject’s head and connected to a Sony digital video camera (model DCR-TRV50). Pre-printed data sheets were used that identified the date, subject, test frequency, sound duration, and speaker locations.

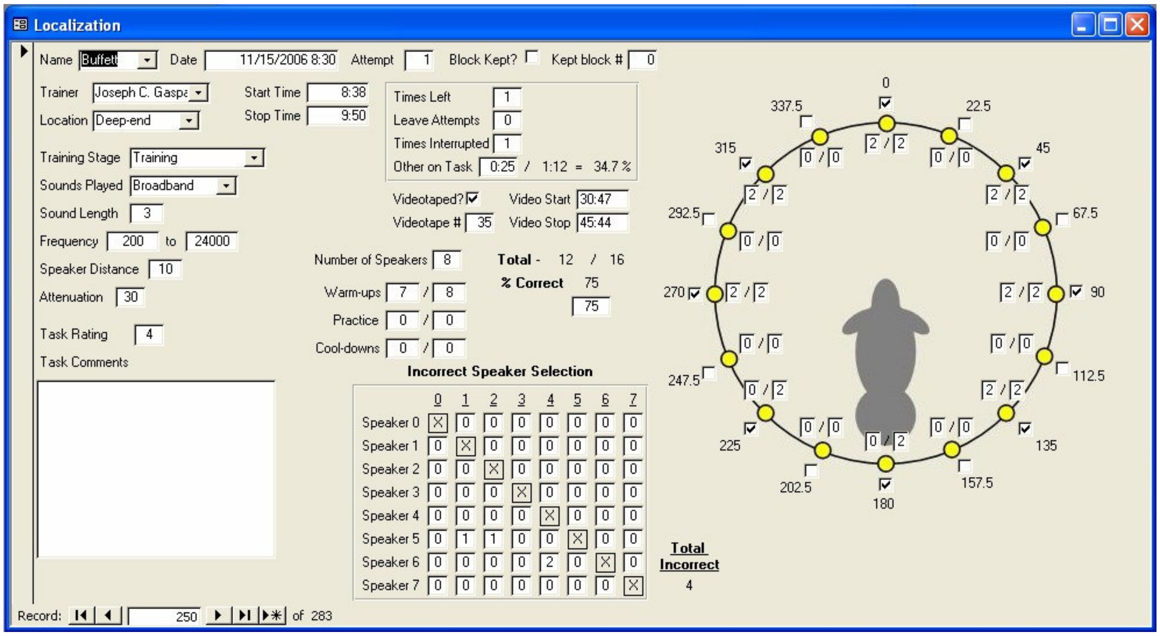

Figure 13. The data entry screen used to enter all of the session's information into the Access database.

Ambient Conditions

All sessions were conducted with the manatee's typical under-water exhibit noise held constant; that is the exhibit's filtration system and pumps ran in their normal capacity. The exhibit noise typically ran below 500 Hz. The in-air noise level, however, was considerably louder than was typical of previous studies with these animals. Construction for a new 3-story building, located less than 200 feet from the manatee exhibit, began just prior to the initiation of training for this study causing intermittent noise of different intensities.

Signal and Speaker Artifacts

All signals had a 100 ms rise-fall time to eliminate transients. Signal intensity levels were randomized by +/- 1.5 dB around the test level to minimize the possibility of intensity level cues being used to determine speaker location (i.e. associating a particular level from a particular speaker).

To control for sound artifacts in the 4-speaker array protocol, the speakers were removed from their original speaker holders and re-connected to the speaker holders diagonally across after three blocks had been completed for each condition (i.e. the speaker located at the 90^0 location for the first three blocks of each condition and then rotated to the 315^0 location for the last three blocks of each condition and this pattern was repeated with other two speakers). All five test signals were recorded from each of the four speakers in their different positions via a Reson hydrophone calibrated and analyzed to ensure that speaker artifacts were not present.

For the 8-speaker array a different method was used to control for speaker differences. A speaker frequency response normalization procedure was developed to eliminate the possibility that small differences in the speakers and tank produced signals could be detected by the manatees and used as a cue in sound localization.

Trainer Cues

Several procedures were followed to avoid trainer cuing for the subjects. All personnel were positioned out of the test subject's line of sight except for the Test Trainer. The Test Trainer was required to wear sound-dampening headphones to avoid the possibility of hearing the test signals and was blind to the test signal's location. The Data-recorder was the only individual who knew where each trial's correct test signal location was, and only obtained that knowledge at the initiation of each trial. To avoid cuing the Test Trainer, the Data-recorder remained positioned behind the computer screen and was not visible until after the subject had made his location selection at the end of each trial. At this point the Test Trainer would look towards the Data-recorder and the Data-recorder would move into view to indicate if the subject's choice was correct or wrong.

Motivational Effects

To control for motivational effects, each animal's session was started with eight "warm-up" trials, presented in a randomized order, and ended with four "cool-down" trials, presented in a randomized order. The signal stimulus used for these trials was the same 0.2 – 20 kHz broad-band noise burst used throughout training.

To control for an apparent initial period of confusion that both manatees displayed when changes between frequencies occurred, "practice" trials were completed immediately after the eight warm-up trials were completed. The signal stimulus used for these trials was the same frequency as the stimulus to be tested in that session. It was believed that although the same spectrum level was used for all of the sound conditions tested, the bandwidth varied with the different stimuli, and the loudness of the test sounds may have been perceived as different to the manatees.

Block Criteria

Two specific criteria were defined as reasons to drop a test block. The first was less than 75% correct on warm-up trials. Blocks were also dropped due to interruptions by the non-tested manatee or frequent departures by the test subject. If a block was dropped, the experimental condition was repeated in the next session.

Testing Trial Sequence

In summary, the sequence of steps utilized for each test trial was as follows:

1) The subject was called to station by the manatee's unique stationing sound originating from the stationing speaker.
2) The subject aligned himself perpendicular to the station bar and pressed the crease of his rostrum against it.

3) The Test Trainer, after ensuring that a 25 second inter-trial interval had passed and that the subject was correctly positioned, told the Data-recorder to play the test sound by verbally stating "tone".
4) The test sound was played from one of the test speakers.
5) The subject swam to and pressed a speaker.
6) The Test Trainer told the Data-recorder the subject's selection.
7) The Data-recorder determined if the speaker location selected matched the actual location from which the test sound was generated.
    - If correct, the subject was presented with the secondary reinforcer sound followed by a food reward. The Test Trainer waited for the animal to stop chewing before initiating the next trial.
    - If wrong, the subject was called back to station and after a 25 sec ITI, the next trial would be initiated.

Results

4-Speaker Experiments

A total of 1,164 trials were run with Hugh and 1,116 trials were run with Buffett. Nine blocks with Buffett and 13 blocks with Hugh were dropped for failure to meet criteria. A total of 60 blocks or 1,008 test trials were kept for each subject.

Five data analyses were conducted for each subject. Two analyses, performance by speaker location and speaker calibration, examined the possibility of the existence of speaker artifactual cues. Analyses for overall performance accuracy, progression learning, and error distribution measured the subject's capacity to localize the test signals.

Speaker Calibration

All five test signals, recorded via a hydrophone located at the center of the stationing bar, were recorded from each of the four speakers when they were positioned in their original locations (speaker 0 was located at 90^0) and again when they were re-positioned and connected to the speaker holders diagonally across (speaker 0 was located at 315^0). The speakers were switched a total of five times throughout testing. This included once after the first three testing blocks of each frequency condition was completed at the first duration condition, a second time after the final three blocks at the first duration and first three blocks at the second duration were complete, a third time when the final three blocks of the second duration and first three of the third duration were complete, a fourth time when the final three blocks of the third duration and first three first three blocks of the fourth duration were complete, and finally a fifth time to complete the last 3 blocks of all frequencies in the fourth duration.

Power spectra were made of all the recordings and examined to look for any frequency or intensity cues that might occur at either a specific location (Figure 14) or from a specific speaker (Figure 15). No obvious patterns were observed, and on the contrary, re-location of the speakers produced only minor signal variations by changing the sound field slightly.

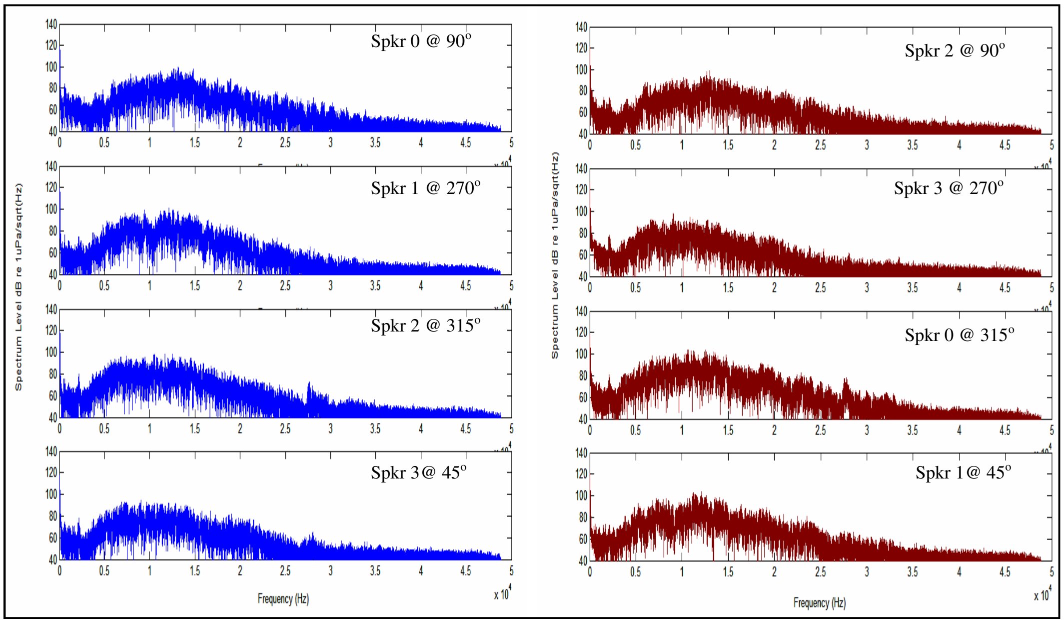

Figure 14. A power spectra comparison of the 6 - 20 kHz broadband test signal from each of the four locations when the speakers were in their original positions (shown on the left in blue) and when they had been re-located shown on the right in red).

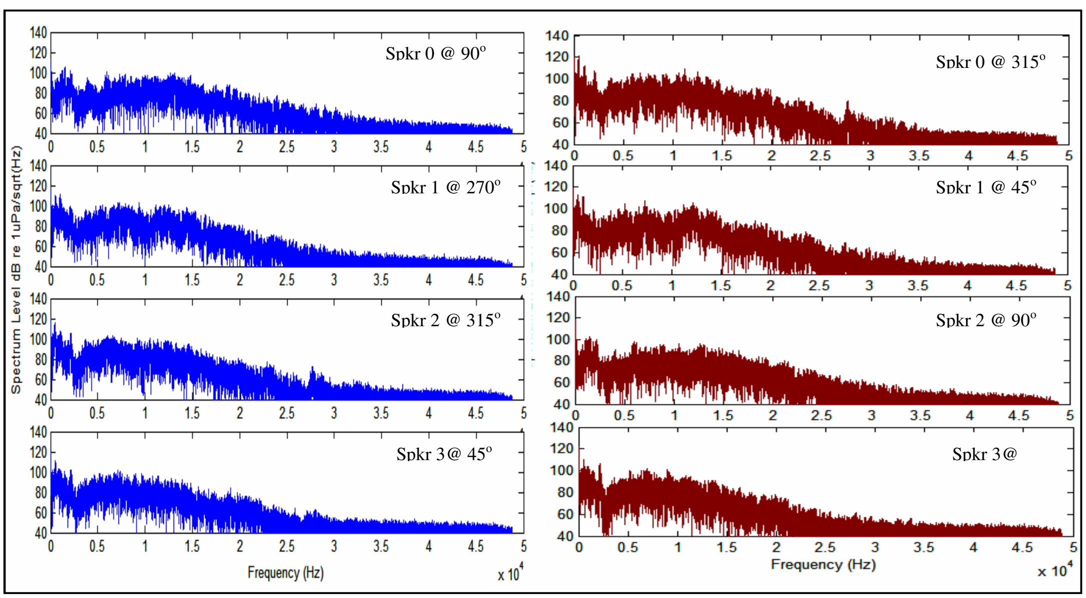

Figure 15. A power spectra comparison of the 0.2 - 20 kHz broadband test signal from each of the four speakers when they were in their original positions (shown on the left in blue) and when they had been re-located (shown on the right in red).

In addition, spectrograms of all the recordings were examined for temporal cues, such as intensity distortions, that might occur within the specific frequencies tested. No obvious patterns or harmonic distortions were observed with either the broad-band

(Figure 16) or pure tone signals (Figure 17). Interestingly, the construction hammering can be observed in the pure tone spectrograms.

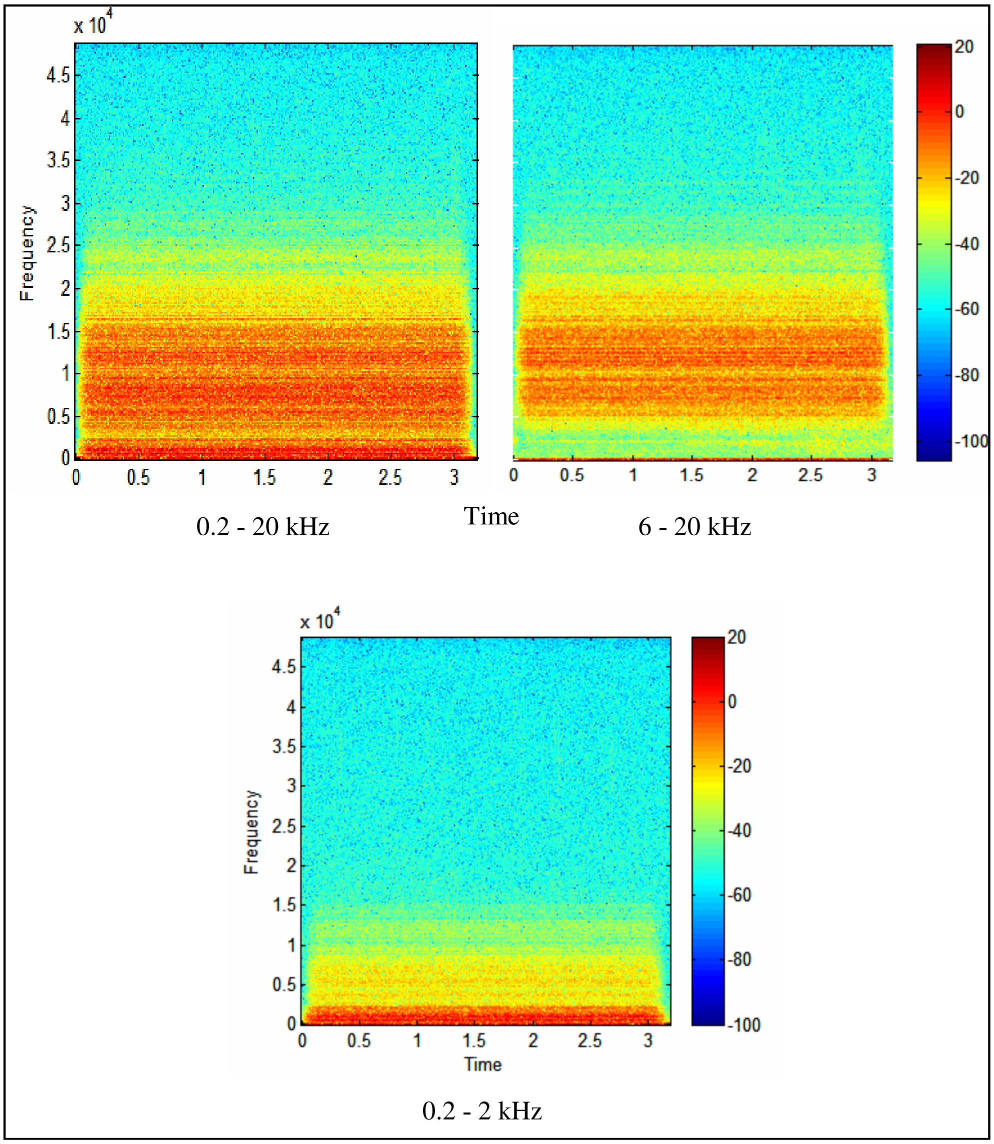

Figure 16. Spectrogram comparison of the three broadband test signals played at the 3000 ms duration (sample rate of 97,656 Hz).

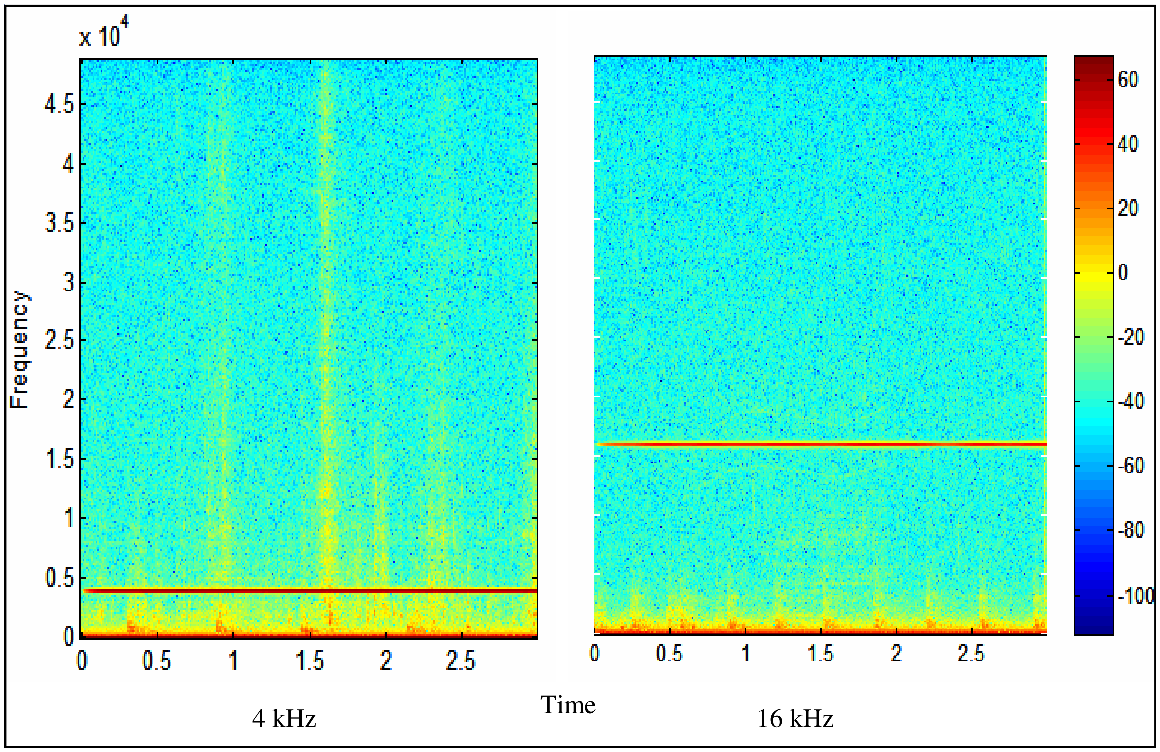

Figure 17. Spectrogram comparison of the two pure tone test signals played at the 3000 ms duration. The recurrent frequency spikes are from the nearby construction hammering (sample rate of 97,656 Hz).

Overall Performance

Overall performance accuracy was determined and described in Table 2. Percentage correct was calculated for each subject based upon 72 trials per condition with a total of 1,008 trials per subject. Both subjects performed well above the 25% chance level for all of the broad-band frequency conditions. Hugh showed a drop in percentage correct as the broad-band signal durations decreased, but this result was not observed with Buffett. Both animals also performed above chance levels with the pure tone signals, but at a much lower accuracy rate than with the broad-band signals.

Table 2
Overall Accuracy Performance per Subject by Frequency and Duration Conditions

	Frequency (kHz)				
Duration:	0.2 - 20	6 - 20	0.2 - 2	4	16
			Hugh		
3000 ms	93%	86%	81%	49%	32%
1000 ms	74%	71%	65%		
500 ms	71%	63%	57%		
200 ms	64%	51%	58%		
			Buffett		
3000 ms	88%	82%	92%	44%	33%
1000 ms	93%	79%	92%		
500 ms	85%	92%	86%		
200 ms	93%	89%	85%		

Note. The values are based on 72 trials per condition with a total of 1,008 trials per subject.

Stability of Response

In order for sensory levels to be accurately measured, performance must be stable across trials. Improved performance associated with learning can render interpretation of response thresholds problematic. Learning was assessed for each subject by comparing the percent correct for each of the six test blocks as they progressed within each of the 14 test conditions (Figure 18). Learning did not appear to occur with Hugh as the individual block accuracy rates had no particular pattern. Buffett demonstrated similar results for all conditions except the broad-band signals presented at the 200 ms duration, where some improvement was observed in the 0.2 - 20 and 6 – 20 kHz conditions.

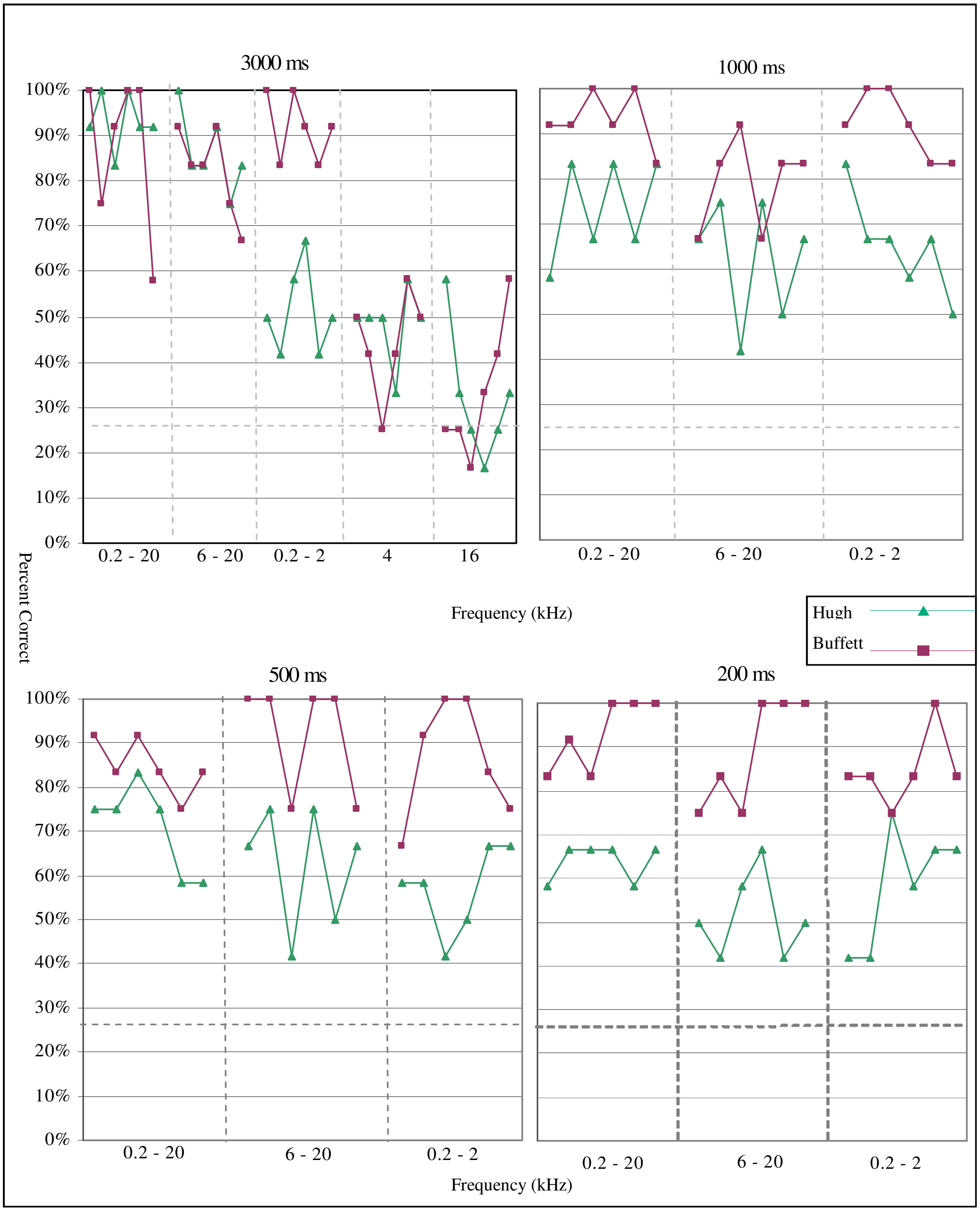

Figure 18. Percent correct by duration for each block as testing progressed across each frequency stimulus.

Error Distribution

The overall error rate, determined from the complete data set collapsed across all conditions, was 29% for Hugh and 19% for Buffett. The broad-band signal error rate, determined from the same data set excluding the tonal signals, was 22% for Hugh and 11% for Buffett.

The distribution of errors made by each subject was examined. An overall percent correct and error distribution was determined, however because the subjects' performance accuracy was considerably lower with pure tone signals, these data were only derived from the results of the broad-band signal testing (Figure 19). Although both subjects had differences in performance accuracy, their distributions of location selection were spatially symmetrical.

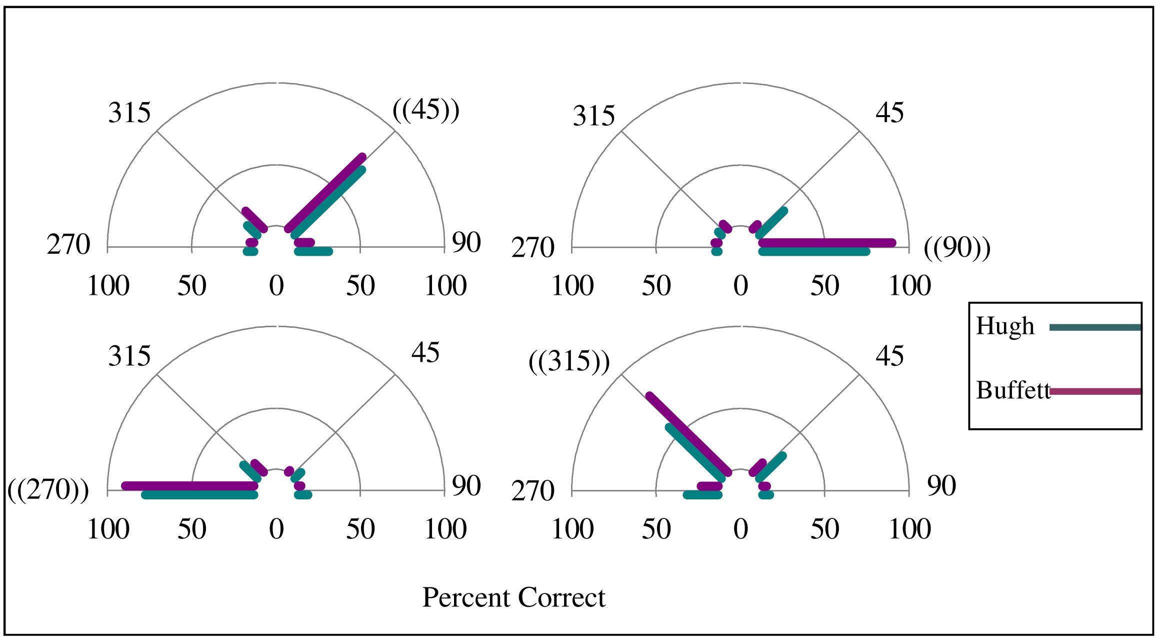

Figure 19. Overall percent correct and distribution of errors using only the results from testing with the broad-band signals. The correct speaker location is notated by double parentheses. Hugh's results are always presented below the graph lines in teal and Buffett's are above the lines in purple.

Error distributions for each tested frequency were determined by comparing the percent correct and incorrect at each speaker location for each of the frequency conditions (Figure 20). The durations were collapsed across the broad-band frequencies however the tonal signals were only tested at the 3000 ms duration. Although both subjects had differences in performance accuracy with the broad-band signals, their errors were generally consistent, with most equally distributed to the locations adjacent to the correct location. For the pure tone signals, errors were scattered among the locations.

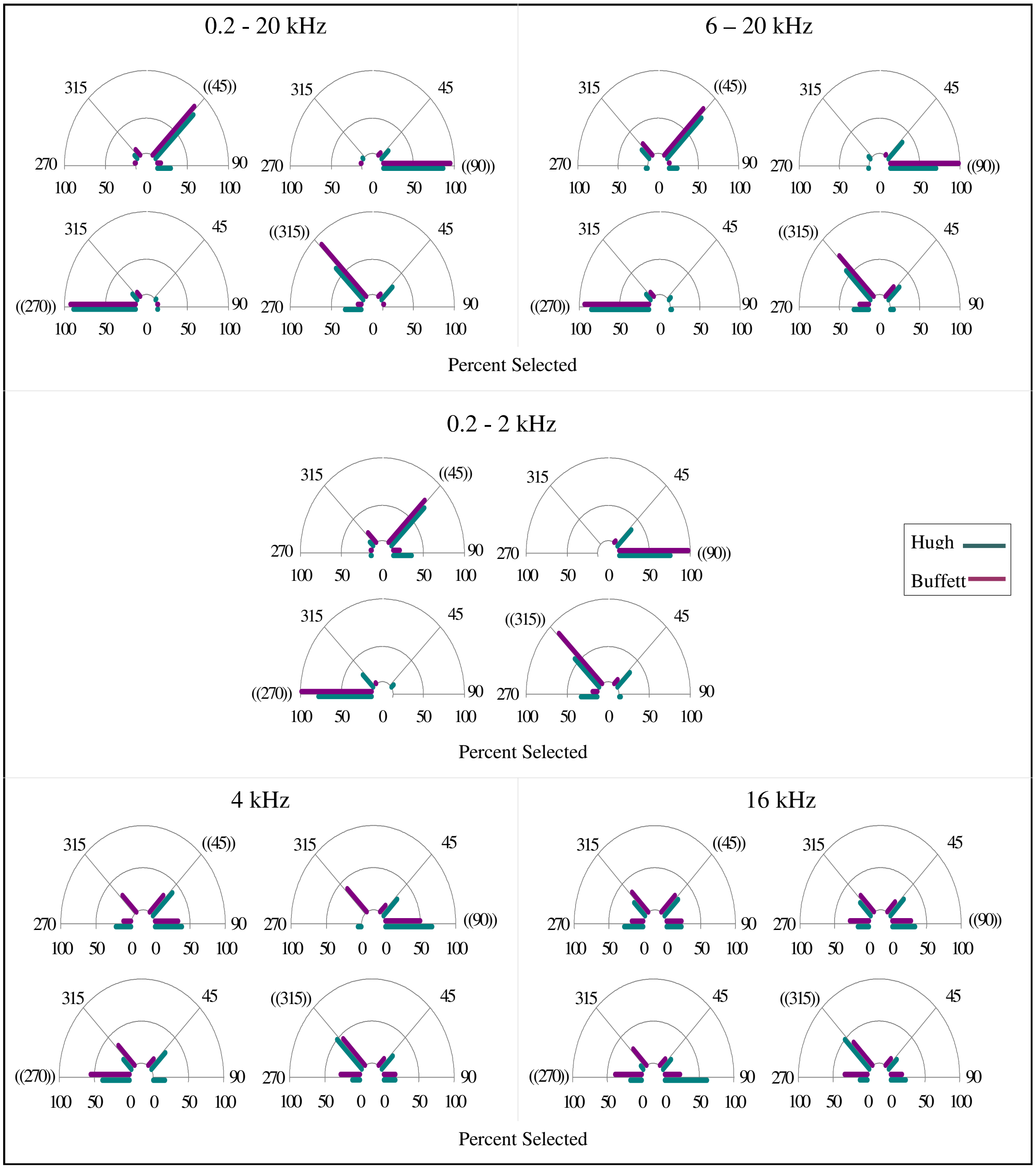

Figure 20. Percent correct and distribution of errors by frequency. The durations were collapsed across the broad-band conditions (top two rows). Tonal conditions are presented in the bottom row. The correct speaker location is notated by double parentheses. Hugh's results are always presented below the graph lines in teal and Buffett's are above the lines in purple.

Error distributions for each sound duration tested were determined by comparing the percent correct and wrong at each speaker location for each of the duration conditions collapsed across the broad-band frequencies (Figure 21). As with the frequency error distributions, the subjects' errors were equally distributed at the locations adjacent to the correct location. Similar results were found when an error distribution compared the percent correct and wrong at each speaker location within the 12 individual broad-band conditions (see Appendix B for all error distributions).

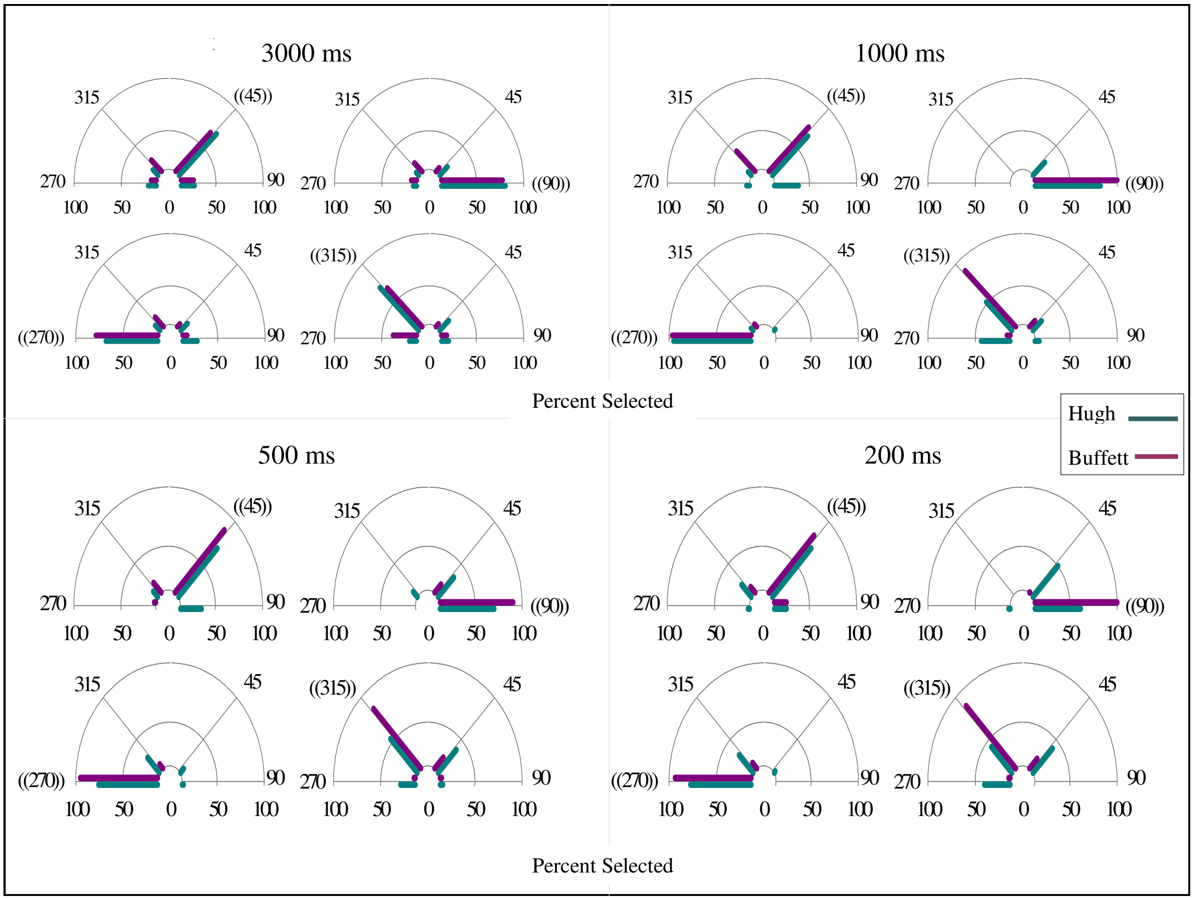

Figure 21. Percent correct and distribution of errors by duration using only the results from testing with the broad-band signals. The correct speaker location is notated by double parentheses. Hugh's results are always presented below the graph lines in teal and Buffett's are above the lines in purple.

8-Speaker Experiments

Speaker Calibration

For the 8-speaker study a speaker frequency response normalization procedure was developed to eliminate the possibility that small differences in the speakers and tank produced signals could be detected by the manatees and used as a cue in sound localization (Fig. 22). This was accomplished by measuring the frequency response of each speaker at the location of the stationing apparatus. Then 500-tap FIR filters were developed for each speaker to produce a flat response over the frequency band. Note that the frequency response is not flat, but louder at lower sound frequencies, which is similar to the spectra produced by boats. The signal also tracks the manatee audiogram, which is more sensitive at higher frequencies (Gerstein et al., 1999).

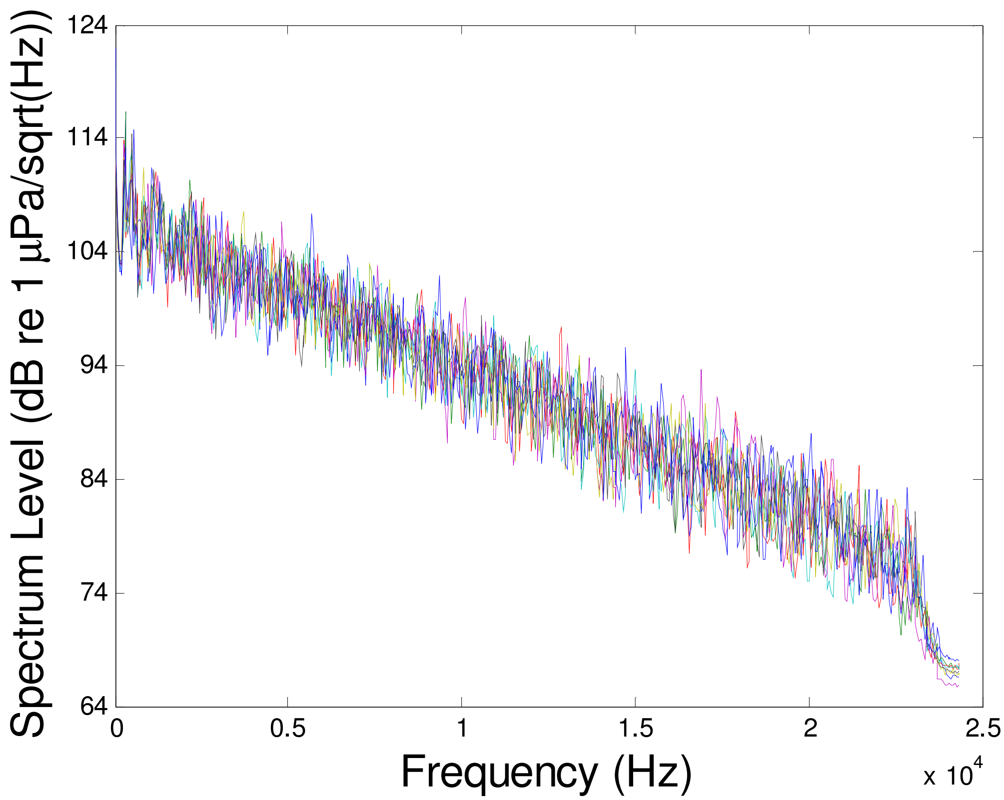

Figure 22. Calibration of sound in 8-speaker experiment. The sound from each speaker was normalized to approximately follow the shape of the manatee audiogram, with decreasing sound levels at higher frequencies. Each curve shows the recording from one of the eight speakers. The speakers are normalized with a 500-tap FIR filter.

Each trial was presented with a sound level that was randomly presented +/- 1.5 dB over the nominal acoustic pressure. There were no large, consistent differences in localization performance prior to and after normalization of the speakers, suggesting that the manatees were not using other cues for sound localization (Table 3).

Overall Performance

Table 4 shows the performance of Hugh and Buffett in the 8-speaker localization experiment. While both manatees performed well above chance, Buffett showed better performance than Hugh. Starting at 125 dB re 1 μPa rms sound level was decreased in 3 dB steps until performance dropped to approximately 50% (well above the 12.5% chance level). For Hugh this was at 15 dB attenuation (110 dB re 1 μPa rms) and for Buffett at 39 dB attenuation (86 dB re 1 μPa rms). Thus Buffett performed better than Hugh at the quieter sound levels tested. Most errors were made by responding to adjacent speakers, with a bias towards the left. Testing of a higher frequency band of noise with Hugh (18-24 kHz) resulted in greatly reduced performance for localization of sounds coming from behind him.

Table 3. Performance of Hugh and Buffet prior to ('Before') and after ('After') speaker normalization. Note that the speaker at 180 degrees is biased by multiple presentations during the 'Before' calibration. 0 degrees is directly in front of the manatee. Data are from training during the summer of 2006.

Speaker (by degree)	***Hugh***		***Buffett***	
	Before	After	Before	After
0	67	50	90	95
45	50	83	95	100
90	50	67	67	95
135	100	83	70	75
180	83	17	50	15
225	17	50	80	80
270	67	67	89	80
315	33	83	70	90

Table 4. Results of 8-speaker localization experiment with Hugh and Buffett. The tables below show percent response for each test condition for each speaker. The speaker that the stimulus was presented from is shown in the first column of the table and the speaker that the manatee chose is shown in the first row of the table. The attenuation refers to sound attenuation relative to 125 dB re 1μPa rms. "Old Speakers" and "New Speakers" refer to the old and new models. All of the speakers were updated to the new model for formal testing.

Hugh

Localization - 20 Blocks (200 - 24,000 Hz, 3 sec, 0 dB Atten.) - Old Speakers

	0	45	90	135	180	225	270	315
Speaker @ 0	73	8	0	0	0	0	5	15
Speaker @ 45	3	93	5	0	0	0	0	0
Speaker @ 90	0	5	73	20	0	0	0	3
Speaker @ 135	3	5	25	60	3	0	5	0
Speaker @ 180	18	10	0	20	10	25	8	10
Speaker @ 225	5	0	5	0	0	60	20	10
Speaker @ 270	0	0	0	3	0	20	50	28
Speaker @ 315	13	0	0	0	0	3	3	83

Localization - 9 Blocks (200 - 24,000 Hz, 3 sec, 0 dB Atten.) - New Speakers

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	6	89	6	0	0	0	0
Speaker @ 135	0	0	6	94	0	0	0	0
Speaker @ 180	17	17	0	17	11	33	6	0
Speaker @ 225	11	0	0	6	0	67	17	0
Speaker @ 270	0	0	0	0	0	39	50	11
Speaker @ 315	11	0	0	0	0	6	6	78

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 3 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	25	75	0	0	0	0	0	0
Speaker @ 90	0	25	75	0	0	0	0	0
Speaker @ 135	0	0	50	50	0	0	0	0
Speaker @ 180	0	0	0	25	25	50	0	0
Speaker @ 225	0	0	0	0	0	75	25	0
Speaker @ 270	0	0	0	0	0	25	50	25
Speaker @ 315	0	0	0	0	0	0	25	75

Localization - 1 Block (200 - 24,000 Hz, 3 sec, 6 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	50	50	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	100	0	0	0	0	0	0	0
Speaker @ 225	0	0	0	0	0	50	50	0
Speaker @ 270	0	0	0	0	50	0	0	50
Speaker @ 315	50	0	0	0	0	0	0	50

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 7 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	75	25	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	50	50	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	0	0	25	75	0	0	0	0
Speaker @ 225	25	0	0	0	0	25	25	25
Speaker @ 270	0	0	0	0	0	0	0	100
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 7 Blocks (200 - 24,000 Hz, 3 sec, 9 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	93	0	0	0	0	0	0	7
Speaker @ 45	14	86	0	0	0	0	0	0
Speaker @ 90	0	14	79	7	0	0	0	0
Speaker @ 135	0	0	21	79	0	0	0	0
Speaker @ 180	14	7	7	14	0	29	7	21
Speaker @ 225	7	0	0	7	0	50	36	0
Speaker @ 270	0	0	0	0	0	7	79	14
Speaker @ 315	0	0	0	0	0	0	14	86

Localization - 1 Block (200 - 24,000 Hz, 3 sec, 12 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	50	50	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	0	0	0	0	0	50	50	0
Speaker @ 225	0	0	0	0	0	50	50	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 5 Blocks (200 - 24,000 Hz, 3 sec, 15 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	60	0	0	0	0	0	10	30
Speaker @ 45	30	50	0	0	0	20	0	0
Speaker @ 90	0	10	50	40	0	0	0	0
Speaker @ 135	0	0	0	90	0	10	0	0
Speaker @ 180	20	0	10	30	0	40	0	0
Speaker @ 225	0	0	10	30	10	30	20	0
Speaker @ 270	0	0	0	10	0	10	30	50
Speaker @ 315	20	0	0	0	0	0	10	70

Localization - 6 Blocks (18,000 - 24,000 Hz, 3 sec, 0 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	75	8	0	0	0	8	8	0
Speaker @ 45	8	92	0	0	0	0	0	0
Speaker @ 90	0	17	58	25	0	0	0	0
Speaker @ 135	8	8	33	33	0	0	17	0
Speaker @ 180	8	8	8	8	0	0	50	17
Speaker @ 225	0	17	8	0	0	0	50	25
Speaker @ 270	8	17	8	8	0	8	33	17
Speaker @ 315	25	0	0	0	0	8	25	42

Buffett

Localization - 27 Blocks (200 - 24,000 Hz, 3 sec, 0 dB Atten.) - Old Speakers

	0	45	90	135	180	225	270	315
Speaker @ 0	94	0	0	0	2	0	0	4
Speaker @ 45	2	96	2	0	0	0	0	0
Speaker @ 90	0	2	80	15	2	2	0	0
Speaker @ 135	0	2	9	61	9	17	2	0
Speaker @ 180	2	4	6	20	17	43	9	0
Speaker @ 225	0	0	0	0	4	83	13	0
Speaker @ 270	0	0	0	2	0	9	87	2
Speaker @ 315	0	2	0	0	0	0	7	91

Localization - 8 Blocks (200 - 24,000 Hz, 3 sec, 0 dB Atten.) - New Speakers

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	88	6	0	0	6	0
Speaker @ 135	6	0	6	56	13	13	6	0
Speaker @ 180	6	0	6	13	63	6	6	0
Speaker @ 225	0	0	0	6	13	81	0	0
Speaker @ 270	6	0	0	0	0	38	56	0
Speaker @ 315	6	0	0	0	0	6	13	75

Localization - 6 Blocks (200 - 24,000 Hz, 3 sec, 5 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	17	83	0	0	0	0
Speaker @ 180	0	0	0	33	67	0	0	0
Speaker @ 225	0	0	0	0	83	17	0	0
Speaker @ 270	0	0	0	0	33	33	33	0
Speaker @ 315	0	0	0	0	17	0	0	83

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 6 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	0	0	0	0	100	0	0	0
Speaker @ 225	0	0	0	0	25	75	0	0
Speaker @ 270	0	0	0	0	0	0	75	25
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 9 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	25	50	0	0	0	25	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	75	0	25	0	0
Speaker @ 180	0	0	0	0	50	50	0	0
Speaker @ 225	0	0	0	0	25	75	0	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 1 Block (200 - 24,000 Hz, 3 sec, 12 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	50	50	0	0	0
Speaker @ 180	0	0	0	50	0	0	50	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 15 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	75	25	0	0	0	0	0
Speaker @ 90	0	0	75	25	0	0	0	0
Speaker @ 135	0	0	0	25	50	25	0	0
Speaker @ 180	0	0	0	25	50	0	25	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 18 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	75	25	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	50	50	0	0	0
Speaker @ 180	0	0	0	0	75	0	25	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	25	75	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 21 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	75	0	0	0	0	0	25	0
Speaker @ 45	0	75	0	0	0	0	25	0
Speaker @ 90	0	0	75	0	0	25	0	0
Speaker @ 135	0	0	0	50	25	25	0	0
Speaker @ 180	0	0	0	0	25	75	0	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 24 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	75	25	0	0	0	0
Speaker @ 135	0	0	25	50	25	0	0	0
Speaker @ 180	0	0	0	0	50	50	0	0
Speaker @ 225	0	0	0	0	25	50	25	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 27 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	50	0	0	50	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	0	75	0	25	0
Speaker @ 180	0	0	0	25	50	25	0	0
Speaker @ 225	0	0	0	0	25	50	25	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Block (200 - 24,000 Hz, 3 sec, 30 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	50	0	25	25	0
Speaker @ 180	0	0	0	75	0	0	25	0
Speaker @ 225	0	0	0	0	25	0	50	25
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 1 Block (200 - 24,000 Hz, 3 sec, 33 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	0	0	0	0	0	50	50	0
Speaker @ 225	0	0	0	0	0	0	0	100
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 1 Block (200 - 24,000 Hz, 3 sec, 36 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	0	100	0	0	0	0
Speaker @ 180	0	0	50	50	0	0	0	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	0	100

Localization - 2 Blocks (200 - 24,000 Hz, 3 sec, 39 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	0	100	0	0	0	0	0	0
Speaker @ 90	0	0	100	0	0	0	0	0
Speaker @ 135	0	0	25	50	0	0	25	0
Speaker @ 180	0	0	0	25	0	25	50	0
Speaker @ 225	0	0	0	0	0	50	50	0
Speaker @ 270	0	0	0	0	0	0	100	0
Speaker @ 315	0	0	0	0	0	0	25	75

Localization - 5 Blocks (200 - 24,000 Hz, 3 sec, 40 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	20	70	0	0	0	0	10	0
Speaker @ 90	0	50	50	0	0	0	0	0
Speaker @ 135	0	0	30	30	20	10	0	10
Speaker @ 180	0	0	10	20	10	30	30	0
Speaker @ 225	0	0	0	0	20	60	20	0
Speaker @ 270	0	0	0	0	10	10	80	0
Speaker @ 315	0	0	0	0	0	10	40	50

Localization - 5 Blocks (200 - 24,000 Hz, 3 sec, 20 dB Atten.)

	0	45	90	135	180	225	270	315
Speaker @ 0	100	0	0	0	0	0	0	0
Speaker @ 45	10	90	0	0	0	0	0	0
Speaker @ 90	0	0	90	0	0	0	10	0
Speaker @ 135	0	0	20	50	20	10	0	0
Speaker @ 180	0	0	10	0	20	70	0	0
Speaker @ 225	0	0	0	0	0	100	0	0
Speaker @ 270	0	0	0	0	0	10	80	10
Speaker @ 315	0	0	0	0	0	0	20	80

Discussion

4-Speaker Experiments

The subjects were able to learn all aspects of the task in the training phases described. The training portion of the study took approximately six months to complete, longer than expected due to some initial technical problems with the computer programming, the start of the manatee's spring mating and migratory period when they are sexually preoccupied and have a tendency to swim stereotypically, and the presence of a sea turtle temporarily residing in the manatee's exhibit. Once these issues were resolved, training was completed rapidly.

The testing portion of the study was completed in approximately six weeks. The subjects were able to localize all of the signals specified within the conditions. These results are not consistent with the contention that manatees are poor at sound localization (cf. Ketten, 1992). Ketten measured the interaural time distance of the manatee and found that they overlapped with the pocket gopher, which was found to be incapable of sound localization (Heffner & Heffner, 1992a). Heffner (1997) has since asserted that head size is not a good predictor of sound localization abilities, and since the results of this study and Gerstein's (1999) localization study indicate that manatees can localize, other considerations of how this is accomplished should be examined.

We found that performance accuracy was higher for broad-band signals than for tonal signals. At 3000 ms duration, Buffett ranged between 92% and 88% correct for broad-band signals, but dropped to 44% and 33% accuracy for tonal signals, likewise, Hugh ranged between 93% and 81% for broad-band signals and 49% and 32% for tonal signals. Because of the low percentage correct, shorter durations for tonal sounds were not tested.

The effect of decreasing sound duration on performance accuracy was mixed. Buffett's overall accuracy was stable (87%, 88%, 88%, and 89%), but Hugh showed a drop in overall accuracy (87%, 70%, 64%, and 58%) as signal duration decreased. The discrepancy between the performances of these two subjects is typical of the results found in other sensory studies that have been conducted with these specific animals (Bauer et al., 2003 & 2005; Mann et al., 2005) and suggest that individual sensory differences are likely to exist. Similarly, substantial individual variability has been found among dolphins who participated in hearing studies (Ridgway & Carder, 1997; Brill et al., 2001)

Both subjects performed above chance levels with the 4 and 16 kHz tonal signals, but at a much lower accuracy rate than with the broad-band signals. The dominant sounds found in the manatee's natural habitat, including boat engine noise, conspecific vocalizations, and ambient noise, are typically composed of numerous broad-band frequencies. These results suggest that although manatees can localize the tonal signals, they are better able to localize the broad-band noises commonly heard in their environment as is typical with most animals (Marler, 1955).

Although the overall error rates within the broad-band conditions were low for both subjects (22% for Hugh and 11% for Buffett), their distribution was consistent and most errors were equally distributed at the locations adjacent to the correct location. Rather than distinguishing a "left vs. right" or "front vs. parallel" strategy, the manatees appeared to use a "nearest neighbor" strategy to localize the broad-band sounds. For the pure tone signals, errors were scattered among the locations and no obvious strategy could be discerned.

8-Speaker Experiments

The 8-speaker experiments showed the manatees were capable of localizing broad-band sounds at all angles, including behind them. This is a challenging task, but especially difficult when the subject can not see the speaker that is producing the test signal. Even with a three second sound duration, the subjects did not have time to turn around to see speakers in back of them before the sound ceased. There were some front-back confusions, as is found with other mammals. However, in a natural situation (such as the approach of a boat) in which the manatee can turn relative to the sound field, these confusions would likely occur less often than in these experiments.

Buffett was able to localize sounds over a fairly large sound level range (95-125 dB re 1µPa rms). However, Hugh's performance dropped at the lower sound levels. This could be the result of a difference in the absolute hearing thresholds of the two manatees, which will be investigated in future studies.

Speaker Artifacts

Numerous controls were put in place to avoid the projection and recognition of speaker artifact cues, including the incorporation of a 100 ms rise-fall time within signals to eliminate transients, the addition of a +/- 1.5 dB randomization within signal levels to eliminate intensity level cues, switching the test signal location during the presentation of the stationing tone, and the routine switching of the speaker locations.

Analysis of the calibration data showed no obvious consistent temporal or harmonic distortions that might be used as cues in the 4-speaker experiments, and were actively filtered in the 8-speaker experiments. In addition, if the subjects used a frequency or speaker artifact cue, a drop in their performance accuracy would be expected between blocks three and four of each condition. For instance, if a particular click was emitted from speaker three and used to identify that speaker's location in the first three blocks, it would be expected that upon switching speaker locations, the subject would continue to select speaker three in its new location and would make incorrect selections. This pattern was not observed and the results suggest that the subjects were localizing the actual test signals and not artifact cues.

Relevance

Results from these experiments have provided information about two manatees' ability to localize specific broad-band and tonal signals of different durations in a controlled environment. This knowledge is important for providing some understanding of how the manatee might detect and localize noise from conspecifics and man-made stimuli such as boats in their natural habitats. Typical recreational boat engines produce broad-band frequencies that range between 0.01 – 2 kHz, although they can reach as high as 20 kHz (Richardson et al., 1995). Manatee vocalizations vary from almost pure tones that tend to modulate between frequencies to broad-band noise and have fundamental frequencies that range from 2.5 – 5.9 kHz but can extend to 15 kHz (Nowacek, et al., 2003). The subjects in this study were able to localize test signals within these same ranges, suggesting that manatees can use localization cues as a means to avoid boats and find conspecifics in their environment.

The knowledge gained through this study could be advanced by additional behavioral research. Testing with Hugh and Buffett was only conducted within the azimuth plane, and future studies might investigate if manatees can also localize sounds within the vertical plane and/or distance dimension. While the results demonstrated that the subjects were able to localize, it is unclear if interaural time, intensity level, and/or phase differences were utilized.

The speaker locations in both experiments were positioned 45^{o} apart. Error distributions suggested that the subjects used a "nearest neighbor" strategy to localize the broad-band test sounds. Almost all of the localization studies conducted with both terrestrial and marine mammals have been minimal audible angle studies, in which the smallest amount of movement of a sound source that can be detected is measured (Mills, 1958). The minimum audible angle study design requires that the subject identify a just detectible change from a particular reference point. The design used in this study required that the subject locate a sound source relative to its own location, which is more difficult, but also relevant to the question of whether manatees can localize sounds.

Understanding how the endangered manatee perceives its environment is a crucial component for making competent management decisions. All of the conservation efforts put in place, including the implementation of boater slow speed zones and manatee preservation areas, have been based upon field studies that identify high manatee abundance areas (Reynolds & Wilcox, 1986) and those that focus on boater behaviors (Gorzelany, 2004). Understanding how the manatee's sensory systems assimilate information and react to environmental stimuli is an important factor that should be considered in conservation management. These studies provide strong evidence that manatees are capable of localizing sounds underwater, including those produced by boats.

References

Ahnelt, P. K. & Kolb, H. (2000). The mammalian photoreceptor mosaic-adaptive design. *Progress in Retinal and Eye Research*, 19, 711-777.

Ames, A. L., Van Vleet, E. S. & Reynolds J. E. (2002). Comparison of lipids in selected tissues of the Florida manatee (Order Sirenia) and bottlenose dolphin (Order Cetacea; Suborder Odontoceti). *Comparative Biochemistry and Physiology*, Part B 132, 625-634.

Anderson, S. (1970). Directional hearing in the harbor porpoise, Phocoena phocoena. In G. Pilleri (Ed.), *Investigations on Cetacea,* Vol II. Berne, Benteli Ag.

Au, W. (1993). *The Sonar of Dolphins*. NY: Springer-Verlag.

Bachteler, D. & Dehnhardt, G. (1999). Active touch performance in the Antillean manatee: Evidence for a functional differentiation of the facial tactile hairs. *Zoology,* 102, 61-69.

Bauer, G. B., Colbert, D. E., Gaspard, J.C., & Littlefield, B., Fellner, W. (2003). Underwater visual acuity of Florida manatees (*Trichechus manatus latirostris*.). *International Journal of Comparative Psychology*, 16, 130-142.

Bauer, G. B., Gaspard III, J. C., Colbert, D.E., Leach, J. B., & Reep, R. (2005, March). *Tactile Discrimination of Textures by Florida Manatees, Trichechus manatus latirostris.* Paper presented at the 12th annual International Conference on Comparative Cognition, Melbourne, Florida.

Brill, R. L., Moore, W. B., & Dankiewicz, L. A. (2001). Assessment of dolphin (*Tursiops truncatus*) auditory sensitivity and hearing loss using jawphones. *Journal of the Acoustical Society of America*, 109, 4, 1717-1722.

Brown, C. H. (1994). Sound Localization. In R. R. Fay and A. N. Popper (Eds.) *Comparative Hearing: Mammals*, New York, Springer-Verlag, pp. 57-97.

Brown, C. H., Beecher, M. D., Moody, D. B., & Stebbins, W. C. (1980). Localization of noise bands by old world monkeys. *Journal of the Acoustical Society of America*, 68, 127-132.

Brown, C. H. & May, B. J. (1990). Sound localization and binaural processes. In M. A. Berkley and W. C. Stebbins (Eds.) *Comparative Perception*, Vol I, New York, John Wiley and Sons, pp. 247-284.

Buckstaff, K. C. (2004). Effects of watercraft noise on the acoustic behavior of bottlenose dolphins, *Tursiops truncatus*, in Sarasota Bay Florida. *Marine Mammal Science*, 20, 709-725.

Bullock, T. H., Domning, D. P., & Best, R. C. (1980). Evoked potentials demonstrate hearing in a manatee (*Trichechus inunguis*). *Journal of Mammalogy*, 61 (1), 130-133.

Bullock, T. H., O'Shea, T. J., & McClune, M. C. (1982). Auditory evoked potentials in the West Indian manatee (Sirenia: *Trichechus manatus*). *Journal of Comparative Physiology*, 148, 547-554.

Burda, H., Bruns, V., Muller, M. (1990). Sensory adaptations in subterranean mammals. In E. Nevo & O. A. Reig (Eds.), *Evolution of Subterranean Mammals at the Organismal and Molecular Levels* (pp. 269-293). NY: Wiley-Liss.

Casseday, J. H. & Neff, W. D. (1973). Localization of pure tones. *Journal of Acoustical Society of America*, 54, 365-372.

Cohen, J. L., Tucker, G. S., & Odell, D. K. (1982). The photoreceptors of the West Indian manatee. *Journal of Morphology,* 173, 197-202.

Colbert, D.E., Fellner, W., Bauer, G. B., Manire, C. & Rhinehart, H. (2001). Husbandry and research training of two Florida manatees, *Trichechus manatus latirostris*. *Aquatic Mammals*, 27 (1), 16-23.

Coren, S., Ward, L. M., and Enns, J. T. (1994). Hearing: Subjective dimensions in sound. In *Sensation and Perception*, 4th Edition, Orlando, Fl, Harcourt Brace & Company, 213-245.

Domjan, M. (1998). *The principles of learning and behavior*. Pacific Grove, CA: Brooks-Cole Publishing Company.

Domning, D. P. & Hayek, L. C. (1986). Interspecific and Intraspecific morphological variation in manatees (Sirenia: Trichechus). *Marine Mammal Science*, 2 (2), 87 - 144.

Don, M. & Starr, A. (1972). Lateralization performance of the squirrel monkey (*Samiri sciureus*) to binaural click signals. *Journal of Neurophysiology*, 35, 493-500.

Fellows, B. J. (1967). Change stimulus sequences for discrimination tasks. *Psychological Bulletin*, 67, 87-92.

Florida Fish & Wildlife Research Institute (2005). *Annual mortality summaries*. http://www.floridamarine.org/features/category_sub.asp?id=2241.

Florida Fish & Wildlife Research Institute (2005). *Annual synoptic surveys*. http://floridamarine.org/features/view_article.asp?id=15246

Gallivan, G. J. & Best, R. C. (1986). The influence of feeding and fasting on the metabolic rate and ventilation of the Amazonian manatee (*Trichechus inunguis*). *Physiological Zoology*, 59 (5), 552-557.

Gellermann, L.W. (1933). Chance orders of alternating stimuli in visual discrimination experiments. *Journal of Genetic Psychology*, 42, 207-208.

Gentry, R.L. (1967). Underwater auditory localization in the California seal lion (*Zalophus californianus*), *Journal of Auditory Research*, 7, 187-193.

Gerstein, E. (1999). *Psychoacoustic Evaluations of the West Indian manatee (Trichechus manatus latirostris).* Unpublished Doctoral Dissertation, Florida Atlantic University, Boca Raton, FL.

Gerstein, E., Gerstein, L., Forsythe, S. & Blue, J. (1999). The underwater audiogram of the West Indian manatee (*Trichechus manatus*). *Journal of the Acoustical Society of America*, 105, 3575-3583.

Gerstein, E. R. (2002). Manatees, bioacoustics and boats. *American Scientist*, 90, 154-163.

Gorzelany, J. F. (2004). Evaluation of boater compliance with manatee speed zones along the Gulf coast of Florida. *Coastal Management*, 32, 215-226.

Griebel, U., & Schmid, A. (1996). Color vision in the manatee (*Trichechus manatus*). *Vision Research,* 36, 2747-2757.

Griebel, U., & Schmid, A. (1997). Brightness discrimination ability in the West Indian manatee (*Trichechus manatus). Journal of Experimental Biology,* 200, 1587-1592.

Hartman, D. S. (1979). *Ecology and Behavior of the manatee (Trichechus Manatus) in Florida.* Special publication No. 5 (153 pp.) The American Society of Marine Mammologists.

Heffner, R. S. (1997). Comparative study of sound localization and its anatomical correlates in mammals. *Acta Otolaryngol*, 532, 46-53.

Heffner, R. S. & Heffner, E. H. (1982). Hearing in the elephant (*Elephas maximus*): Absolute sensitivity, frequency discrimination, and sound localization. *Journal of Comparative and Physiological Psychology,* 96 (6), 926-944.

Heffner, R. S. & Heffner, E. H. (1984). Sound localization in large mammals: Localization of complex sounds by horses. *Behavioral Neuroscience*, 98 (3), 541-555.

Heffner, R. S. & Heffner, E. H. (1985). Sound localization in wild Norway rats (*Rattus Norvegicus*). *Hearing Research*, 19, 151-155.

Heffner, R. S. & Heffner, E. H. (1988). Sound localization acuity in the cat: Effect of azimuth, signal duration and test procedure. *Hearing Research*, 36, 221-232.

Heffner, R. S. & Heffner, E. H. (1988). Sound localization in a predatory rodent, the Northern Grasshopper mouse (*Onychomys leucogaster*). *Journal of Comparative Psychology*, 102 (1), 66-71.

Heffner, R. S. & Heffner, E. H. (1988). Sound localization and use of binaural cues by the gerbil (*Meriones unguiculatus*). *Behavioral Neuroscience*, 102 (3), 422-428.

Heffner, R. S. & Heffner, E. H. (1989). Sound localization, use of binaural cues and the superior olivary complex in pigs. Brain Behavior Evolution, 33, 248-258.

Heffner, R. S. & Heffner, H. E. (1990). Vestigial hearing in a fossorial mammal, the pocket gopher, (*Geomys bursarius*). Hearing Research, 46, 239-252.

Heffner, R. S. & Heffner, E. H. (1992a). Evolution of sound localization in mammals. In, D. Webster, R. Fay and A. Popper (Eds.) *The Biology of Hearing*, Springer-Verlag, pp. 691-715.

Heffner, R. S. & Heffner, E. H. (1992b). Hearing in large mammals: Sound localization acuity in cattle *(<u>Bos Taurus</u>*) and goats *(Capra hircus*). *Journal of Comparative Psychology*, 106 (2), 107-113.

Heffner, R. S. & Masterson, R. B. (1990). Sound localization in mammals: Brain stem mechanisms. In M. A. Berkley, & W. C. Stebbins (Eds.) *Comparative Perception*, New York, John Wiley and Sons, pp. 285-314.

Heinsohn, G. E., Marsh, H., & Spain, A. V. (1976). Extreme risk of mortality to dugongs (Mammalia: Sirenia) from netting operations. *Australian Journal of Wildlife Research*, 3, 117-121.

Holt, M. M., Schusterman, R. J., Southall, B. L., & Kastak, D. (2004). <u>Localization of aerial broadband noise by pinnipeds</u>. *Journal of the Acoustical Society of America*, 115, 2339-2345.

Houben, D. & Gourevitch, G. (1979). Auditory lateralization in monkeys: An examination of two cues serving directional hearing. *Journal of Acoustical Society of America*, 66, 1057-1063.

Isley, T. E. & Gysel, L. W. (1975). Sound source localization by the red fox. *Journal of Mammalogy*, 56, 397-404.

Ketten, D. R., Odell, D. K., Domning, D. P. (1992). Structure, function, and adaptation of the manatee ear. In J. A. Thomas, R. A. Kastelein, & A. Y. Supin (Eds.) *Marine Mammal Sensory Systems* (pp.77-79). NY: Plenum Press.

Kirkpatrick, B., Colbert, D. E., Dalpra, D., Newton, E. A. C., Gaspard, J., Littlefield B., & Manire, C. A. (2002). Florida Red Tides, Manatee Brevetoxicosis, and Lung

Models. In K. A. Steidinger, J. H. Landsberg, C. R. Tomas, and G. A. Vargo (Eds.) *Xth International Conference on Harmful Algae. Harmful Algae*. Florida Fish and Wildlife Conservation Commission and Intergovernmental Oceanographic Commission of UNESCO.

Klishen, V. O., Diaz, R. P., Popov, V. V., & Supin, A. Y. (1990). Some characteristics of hearing of the Brazilian manatee, *Trichechus inunguis*. *Aquatic Mammals*, 16 (3), 139-144.

Manire, C. A., Walsh, C. J., Rhinehart, H. L., Colbert, D. E., Noyes, D. R., & Luer, C. A. (2003). Alterations in blood and urine parameters in two Florida manatees, *Trichechus manatus latirostris*, from simulated conditions of release following rehabilitation. *Zoo Biology*, 22, 103-120.

Mann, D., Colbert, D. E., Gaspard, J. C. III, Casper, B., Cook, M. L. H., Reep, R. L., & Bauer, G. B. (2005). Temporal resolution of the Florida manatee (*Trichechus manatus latirostris*) auditory system. *Journal of Comparative Physiology*, 191, 903-908.

Marler, P. (1955). Some characteristics of animals calls. *Nature*, 176, 6, 6-8.

Marsh, H., Heinsohn, G. E., & Marsh, L. M. (1984). Breeding cycle, life history, and population dynamics of the dugong, *Dugong dugon* (Sirenia: Dugongidae). *Australian Journal of Zoology*, 32, 767-788.

Marshall, C. D., Huth, G. D., Edmonds, V. M. Halin, D. L., & Reep, R. L. (1998). Prehensile use of perioral bristles during feeding and associated behaviors of the Florida manatee (*Trichechus manatus latirostris*). *Marine Mammal Science,* 14, 274-289.

Marshall, C. D., Maeda, H., Iwata, M., Furuta, M. Asano, A., Rosas, F., & Reep, R. L. (2003). Orofacial morphology and feeding behavior of the dugong, Amazonian, West African, and Antillean manatees (Mammalia: Sirenia): Functional morphology of the muscular-vibrissal complex. *Journal of Zoology*, 259, 245-260.

Mass, A. M., Odell, D. K., Ketten, D. R., & Supin, A. Y. (1997). Ganglion layer topography and retinal resolution of the Caribbean manatee *Trichechus manatus latirostris*. *Doklady Biological Sciences,* 355, 392-394.

Masterson, B., Thompson, G. C., Bechtold, J. K. & RoBards, M. J. (1975). Neuroanatomical basis of binaural phase-difference analysis for sound localization: A comparative study. *Journal of Comparative Physiological Psychology*, 89, 379-386.

Medwin, H., & Clay, C. S. (1998). *Fundamentals of Acoustical Oceanography*. NY: Academic Press.

Mills, A. W. (1958). On the minimum audible angle. *Journal of the Acoustical Society of America*, 30, 127-246.

Mills, A. W. (1972). Auditory localization. In J. V. Tobias (Ed.) Foundations of Modern Auditory Theory: Vol. II, (pp. 3030-348). NY: Academic Press

Moore, P. W. B. (1974). Underwater localization of click and pulsed pure-tone signals by the California sea lion (*Zalophus californianus*). *Journal of the Acoustical Society of America*, 57 (2), 406-410.

Moore, P. W. B. & Au, W. L. (1975). Underwater localization of pulsed pure tones by the California sea lion (*Zalophus californianus*). *Journal of the Acoustical Society of America,* 58 (2), 721-727.
Moore, P. W. B. & Brill, R. L. (2001). Binaural hearing in dolphins. *The Journal of the Acoustical Society of America*, 109 (5), 2330-2331.
Moore, P. W. B. and Pawloski, D. A. (1993). Interaural time discrimination in the bottlenose dolphin. *The Journal of the Acoustical Society of America,* 94 (3), 1829-1830.
Nachtigall, P. E. (1985). Vision, audition, and chemoreception in dolphins and other marine mammals. In R.L. Schusterman, J. A. Thomas, & F.G. Wood (Eds.), *Dolphin cognition and behavior: A comparative approach* (pp. 79-113). Hillsdale, NJ: Erlbaum.
Nowacek, D. P., Casper, B. M., Wells, R. W., Nowacek, S. M., & Mann, D. A. (2003). Intraspecific and geographic variation of West Indian manatee (*Trichechus manatus spp*.) vocalizations. *Journal of the Acoustical Society of America*, 114 (1), 66-69.
Nowacek, S. M., Wells, R. S., Owen, E. C. G., Speakman,T. R., Flam, R. O., & Nowacek, D. P. (2004). Florida manatees, *Trichechus manatus latirostris*, respond to approaching vessels. *Biological Conservation* 119, 517-523.
Odell, D. K., & Reynolds, J. E. III. (1979). Observations on manatee mortality in South Florida. *Journal of Wildlife Management*, 28 (3), 572-577.
Pepper, R. L., & Defran, R. H. (1975). *Dolphin Trainer's Handbook, Part 1: Basic Training* (50p). Naval Undersea Center, San Diego, CA.
Piggins, D. J., Muntz, W. R. A., & Best, R. C. (1983). Physical and morphological aspects of the eye of the manatee, *Trichechus inunguis*. *Marine Behaviour and Physiology***,** 9, 111-130.
Popov, V. V. & Supin, A.Y. (1990). Electrophysiological studies on hearing in some cetaceans and a manatee. In J.A. Thomas, & R. A. Kastelein (Eds.), *Sensory Abilities of Cetaceans: Laboratory and Field Evidence*. NY: Plenum Press.
Ramirez, K. (1999). *Animal Training: Successful animal management through positive reinforcement.* Shedd Aquarium, Chicago, Illinois.
Reep, R. L.**,** Marshall, C. D., Stoll, M. L., Homer, B. L. & Samuelson, D. A. (2001). Microanatomy of facial vibrissae in the Florida manatee: the basis for specialized sensory function and oripulation. *Brain Behavior and Evolution*, 58, 1-14.
Reep**,** R. L.**,** Marshall, C. D., & Stoll, M. L. (2002). Tactile hairs on the postcranial body in Florida manatees: a mammalian lateral line? *Brain Behavior and Evolution*, 59, 141-154.
Reeves, R. R., Tuboku-Metzger, D. & Kapindi, R. A. (1988). Distribution and exploitation of manatees in Sierra Leone. *Oryx*, 12 (4), 465-470.
Renaud, D. L., & Popper, A. N. (1975). Sound localization by the bottlenose porpoise *Tursiops truncatus*. *Journal of Experimental Biology*, 63, 569-585.
Reynolds, J. E. III. (1979). The semisocial manatee. *Natural History*, 88 (2), 44-53.
Reynolds, J. E. & Odell, D. K. (1991). *Manatees and Dugongs* (192 pp.). NY: Checkmark Books.

Reynolds, J. E. III, & Wilcox, J. R. (1986). Distribution and abundance of the West Indian manatee, *Trichechus manatus,* around selected Florida power plants following winter cold fronts: 1984-1985. *Biological Conservation*, 38, 103-113.

Richardson, W., Greene, C., Malme, C. & Thompson, D. (1995). *Marine mammals and noise*. San Diego, CA: Academic Press.

Ridgway, S. H. & Carder, D. A. (1997). Hearing deficits measured in some *Tursiops truncatus*, and discovery of a deaf/mute dolphin. *Journal of the Acoustical Society of America*, 101, 590-594.

Ross, D. (1976). *Mechanics of Underwater Noise.* NY: Pergamon.

Smith, A. & Marsh, H. (1990). Management of traditional hunting of dugongs [*Dugong dugon* (Muller, 1976)] in the northern Great Barrier Reef, Australia. *Environmental Management*, 14 (1), 47-55.

Stevens, S. S. & Newman, E. B. (1936). The localization of actual sources of sound. *American Journal of Psychology*. 48, 297-306.

Supin, A. Y., Popov, V. V., & Mass, A. M. (2001). *The sensory physiology of aquatic mammals*. MA: Kluwer Academic Publishers.

Terhune, J. M. (1974). Directional hearing of a harbor seal in air and water. *Journal of the Acoustical Society of America,* 56 (6), 1862-1865.

Timm, R. M., Albuja, L. V. & Clauson, B. L. (1986). Ecology, distribution, harvest, and conservation of the Amazonian manatee, *Trichechus inunguis* in Ecuador. *Biotropica*, 18 (2), 150-156.

Urick, R. J. (1996). *The principles of underwater sound, 3rd Edition* (423pp.). CA: Peninsula Publications.

U.S. Fish and Wildlife Service (2001). *Florida Manatee Recovery Plan, (Trichechus manatus latirostris), Third Revision*. U.S. Fish and Wildlife Service, Atlanta, Georgia. 144 pp.

Wakeford, O. S. and Robinson, D. E. (1974). Lateralization of tonal stimuli by the cat. *Journal of the Acoustical Society of America*, 55, 649-652.

Walls, G. L. (1963). *The vertebrate eye and its adaptive radiation*. NY: Hafner.

West, J. A., Sivak, J. G., Murphy, C. J., & Kovacs (1991). A comparative study of the anatomy of the iris and ciliary body in aquatic mammals. *Canadian Journal of Zoology,* 69, 2594-2607.

Wartzok, D. & Ketten, D. (1999). Marine Mammal Sensory Systems. In J. E. Reynolds & S. A. Rommel (Eds.), *Biology of Marine Mammals* (600 pp.). Smithsonian Institute Press.

Yost, W. A. (2000). Sound localization and binaural hearing. *In Fundamentals of hearing: An introduction* (pp. 179-192). San Diego, CA: Academic Press.

Appendix A

The Computer Protocols Used For all Phases of the Experimental Conditions

A graphical user interface, programmed in Visual C was designed to run each phase of the experimental conditions (Figure A1). A drop-down subject menu was designed to distinguish which subject was being tested, and this selection automatically referenced and played that animal's stationing and reinforcement. A "notes" section allowed any comments to be digitally recorded relative to that block.

The "set-up" section defined how many speaker locations were to be tested, how many trials were to be run from each of those speakers, and how many of the test sounds could be played from the same location in a row. In addition, broad-band noise bursts or tonal signals were defined as were the frequency range, sound duration, dB level, and digital recording.

The "speaker" section provided information about speaker location. If needed, a manual switching check box was included, which allowed the Data-recorder to select the location of the test sound to be played, rather than the randomized location generated by the program.

The "status" section defined and digitally recorded how many trials had been completed within the block, and of those, how many were correct and how many were incorrect. The start button initiated the block of twelve trials once the subject and conditions were defined, and the stop button was used only if the block had to be ended prior to the completion of the twelve trials.

The correct experimental conditions were incorporated for each portion of the session, including the warm-up, practice, testing, and cool-down trials (a total of 56 trials were run per animal per session). In all portions of a session, four speaker locations, a maximum of two trials in a row per location, and a randomized level of three dB were held constant.

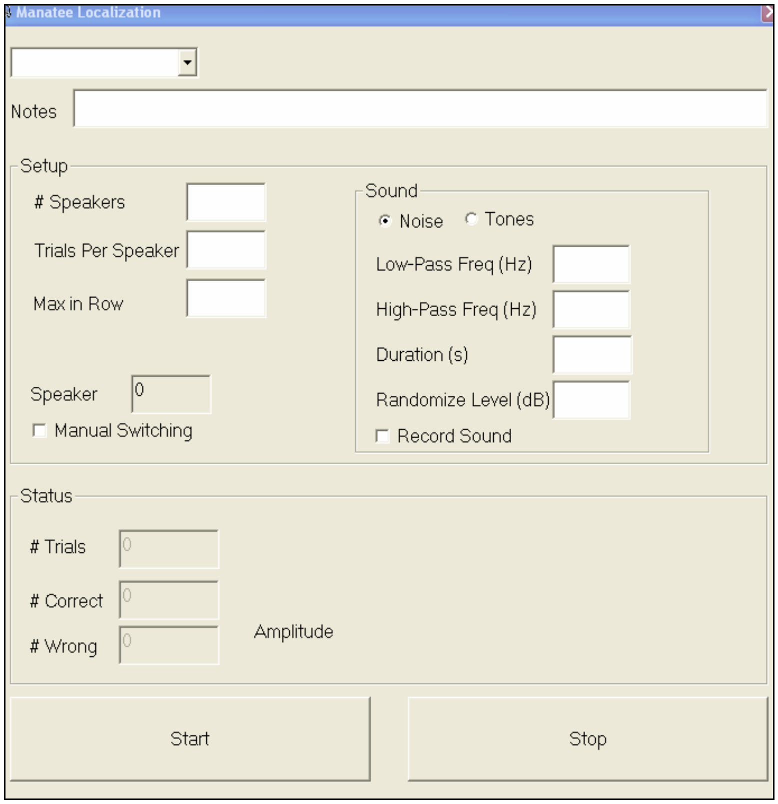

Figure A1: The graphical user interface screen (programmed in Visual C) used to setup the experimental conditions and automatically download the results into an Excel file during the testing sessions.

Appendix B

Percentage correct and error distribution within the 12 broad-band conditions in the 4-speaker experiments. An error distribution was determined by comparing the percent correct to the percent wrong at each speaker location within the 12 individual broad-band conditions. The error distribution is shown for all of the duration conditions within the 0.2 - 20 (Figure B1), 6 - 15 (Figure B2), and 0.2 - 2 kHz (Figure B3) broad frequency conditions.

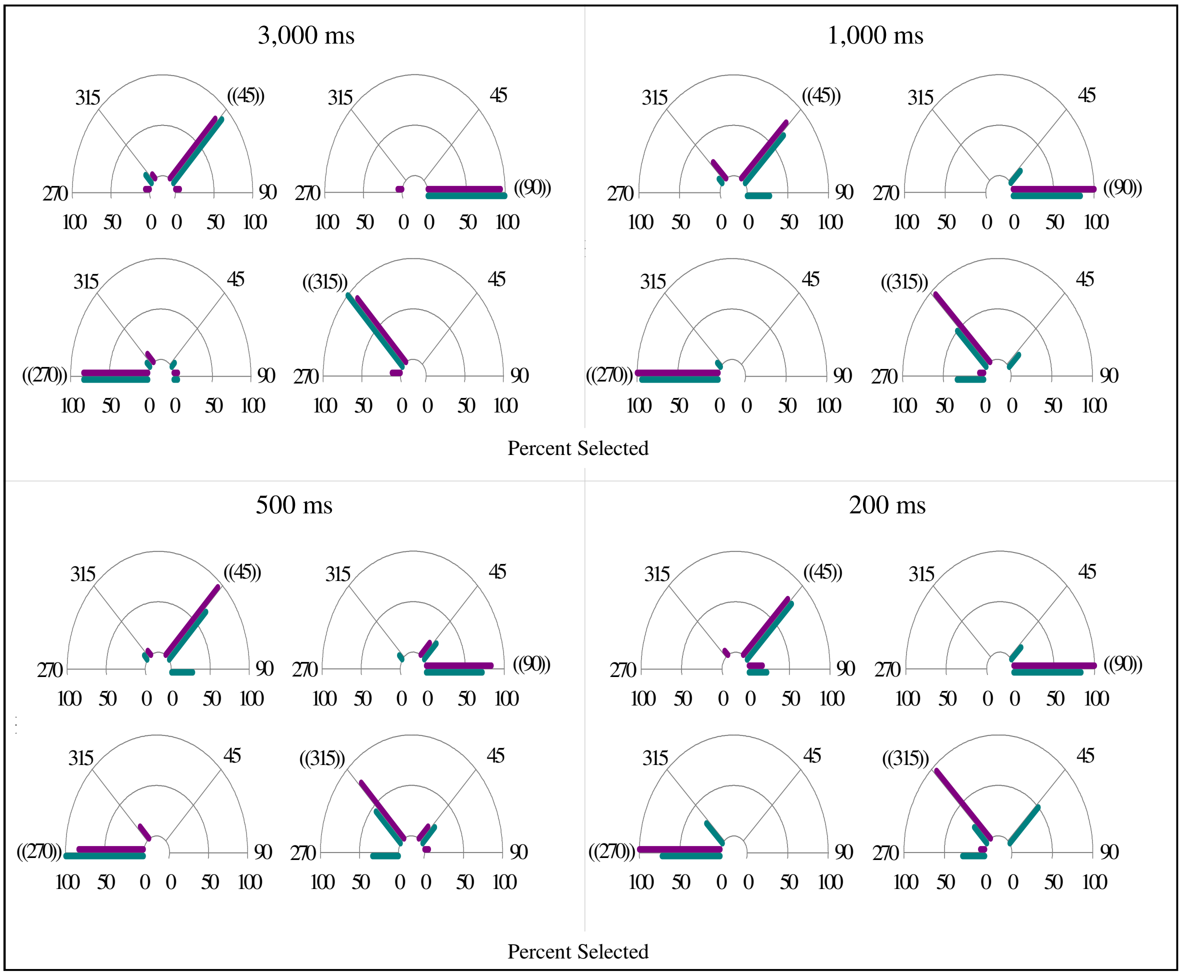

Figure B1. Percent correct and distribution of errors by duration within the 0.2 - 20 kHz condition. The correct speaker location is notated by double parenthesis. Hugh’s results are always presented below the graph lines in teal and Buffett’s are above the lines in purple.

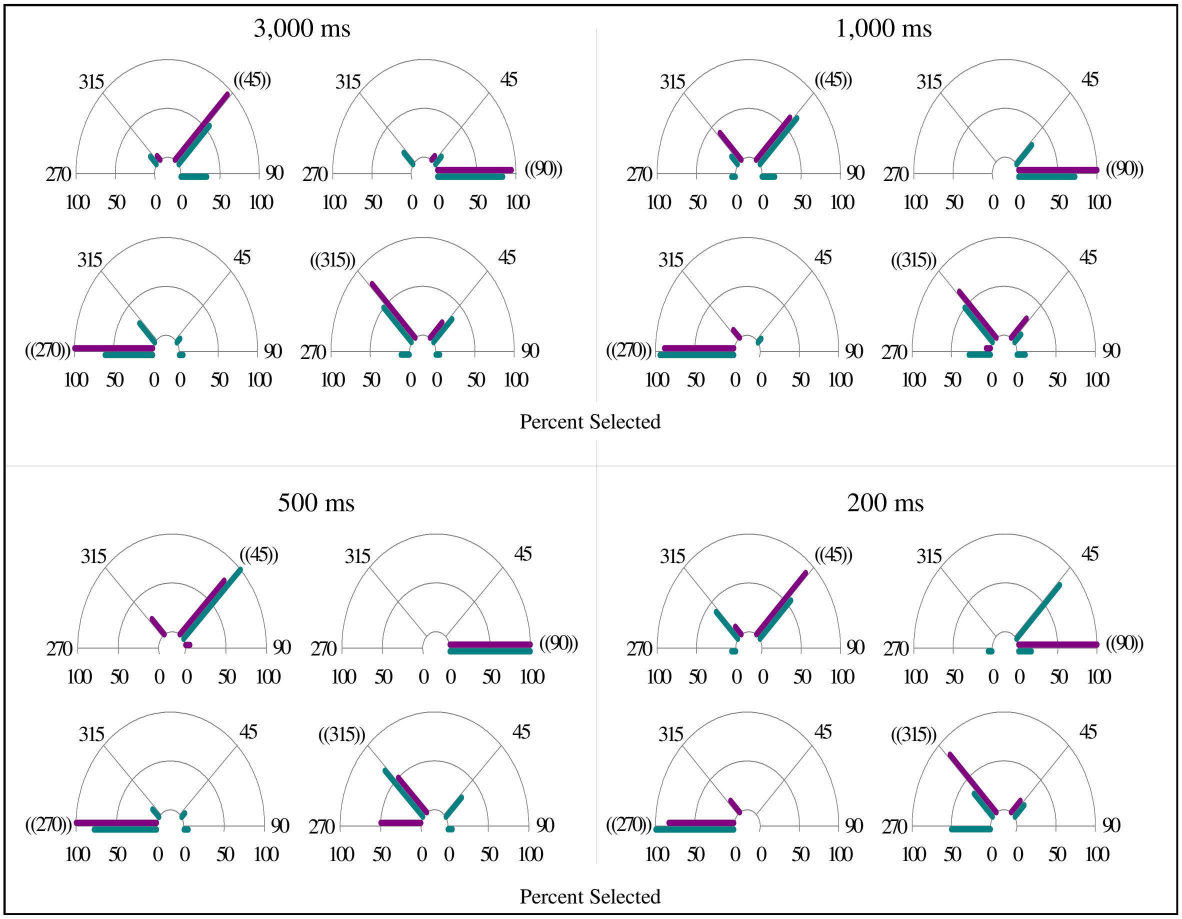

Figure B2. Percent correct and distribution of errors by duration within the 6 - 20 kHz condition. The correct speaker location is notated by double parenthesis. Hugh's results are always presented below the graph lines in teal and Buffett's are above the lines in purple.

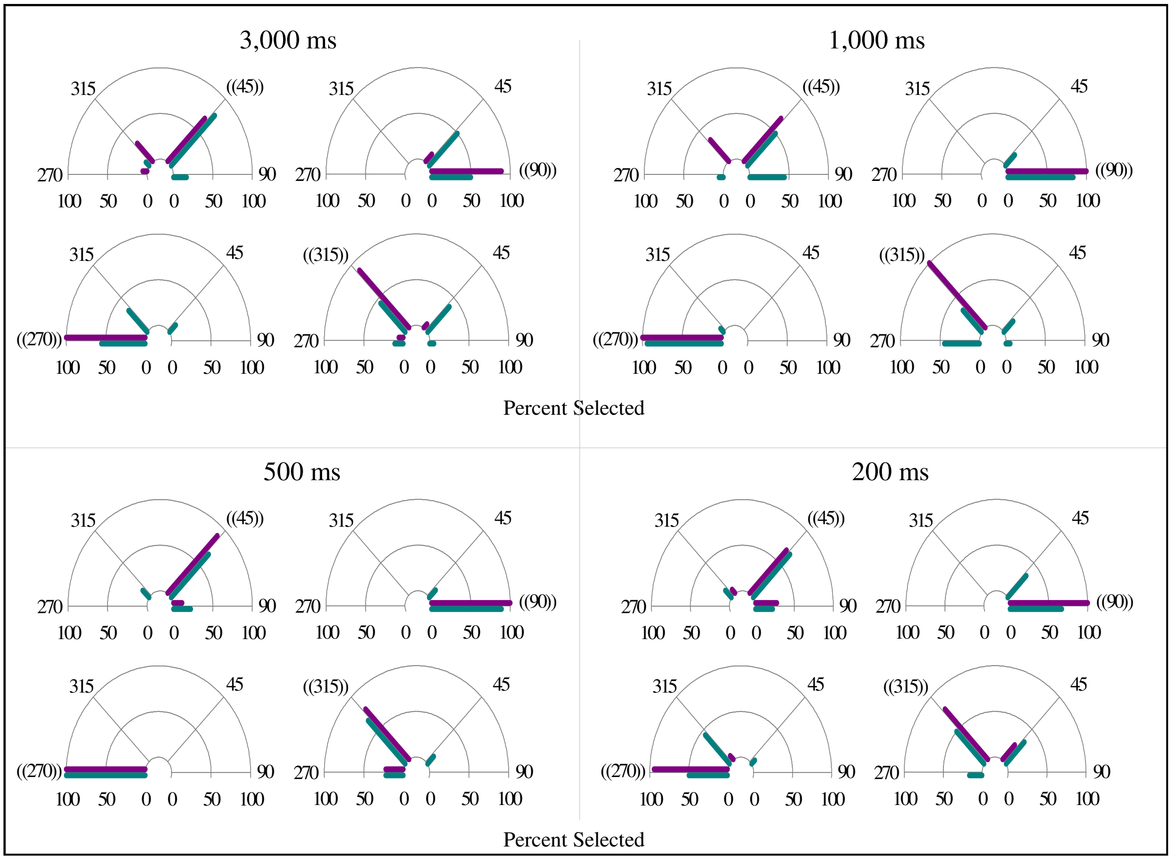

Figure B3. Percent correct and distribution of errors by duration within the 0.2 - 2 kHz condition. The correct speaker location is notated by double parenthesis. Hugh's results are always presented below the graph lines in teal and Buffett's are above the lines in purple.