HEARING THRESHOLDS, FREQUENCY DISCRIMINATION, AND ACOUSTIC ORIENTATION IN THE LEMON SHARK, *NEGAPRION BREVIROSTRIS* (POEY)1

DONALD R. NELSON2

Institute of Marine Science, University of Miami

ABSTRACT

Laboratory experiments are described in which the hearing of young lemon sharks was investigated using several conditioning methods. Hearing-threshold curves were obtained by approach, arousal, and escape conditioning, and are presented in absolute decibels and in decibels relative to ambient noise. Limits of frequency discrimination are given which were based on experiments utilizing approach-avoidance and escape conditioning. Acoustic orientation in an approach-conditioned shark is described and mean orientation errors for several frequencies are given. Theories of hearing in fish are reviewed and comparisons are drawn between present results and those of previous studies. The significance of these results is discussed in the light of present acoustical concepts, such as pressure and displacement, and the near and far fields.

INTRODUCTION

The study of hearing in sharks is of considerable interest both because it relates to the general problem of hearing mechanisms in fish, and because it relates to the special problem of shark attack. It appears that hearing is one of the means by which sharks locate their prey. Field observations by several workers have suggested that sharks utilize sounds to locate struggling fish or other disturbances (Hobson, 1963; Limbaugh, 1963; Randall, 1963; Eibl-Eibesfeldt & Hass, 1959; and Wright, 1948). Experiments by Nelson & Gruber (1963), using low-frequency, pulsed sounds, have confirmed that large sharks in their natural environment can be attracted by sounds alone. Subsequent experiments have shown that sharks can detect, and orient to, these sounds at distances of at least 600 feet (Wisby & Nelson, 1964; Nelson, 1965).

Several review articles have been published concerning the sense of hearing in elasmobranchs. Backus (1963) and Wisby et al. (1964) addressed themselves directly to this subject. Dijkgraaf (1963b) thoroughly reviewed the experimental studies on the function of the lateral-line organs. Reviews of hearing in fish by Moulton (1963) and Lowenstein (1957) included material on elasmobranchs. Recent works of Tavolga & Wodin-

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2 Present address: Department of Biology, California State College at Long Beach, Long Beach, California.
sky (1963) and Tavolga (1965) have brought together much of the pertinent information on hearing in both teleosts and elasmobranchs.

The first experimental studies on the sense of hearing in sharks were those of Parker (1905, 1910) who obtained unconditioned responses from smooth dogfish, *Mustelus canis*, by shaking the whole aquarium or by striking the side of the aquarium with a hammer. Vilstrup (1951) conditioned *Acanthias vulgaris* (= *Squalus acanthias*) to respond to the broad spectrum sound of a motor horn. Unpublished results by Moulton (personal communication) in 1955 indicated that *Mustelus canis* can be conditioned to a 1,000 cps tone. Clark (1959), while establishing instrumental conditioning in large lemon sharks, *Negaprion brevirostris*, observed that the sharks were able to detect and respond to the sound of a submerged bell. Preliminary results by Dijkgraaf (1963a) show conditioned responses to a 180-cps tone by the dogfish, *Scyliorhinus canicula*.

Kritzler & Wood (1961), working with a captive bull shark, *Carcharhinus leucas*, obtained the first complete audiogram for a shark. They trained the shark to approach an underwater loudspeaker to obtain a food reward. The shark responded to frequencies between 100 and 1,500 cps and was most sensitive to the band between 400 and 600 cps, where its threshold was below filtered ambient noise. An additional observation was that the shark was able to localize the sound source from a distance of at least 20 feet (Wood, personal communication).

Olla (1962) trained small hammerhead sharks, *Sphyrna lewini*, to associate sounds with an unconditioned stimulus consisting of a yank on a cord fastened to the dorsal fin. Using both underwater and air speakers, he concluded that the sharks could perceive frequencies over a range of at least 250 to 750 cps with maximum sensitivity between 250 and 375 cps.

Working at the oceanarium in Durban, South Africa, Davies *et al.* (1963) obtained conditioned responses to sounds from a group of young sharks of the following species: dusky sharks, *Carcharhinus obscurus*; black-tip sharks, *C. maculipinnis*; bull sharks, *C. leucas*; and hammerhead sharks, *Sphyra lewini*. Davies *et al.* reported conditioning to pure tones over a frequency range of 50 to 7,000 cps. Accurate directional hearing was also noted. A theory was advanced suggesting that hearing in sharks is velocity (displacement) operated, and that directional hearing is based on directional characteristics of the inner ears.

**Acknowledgments**

This study was part of a continuing investigation of the behavior and sensory physiology of sharks currently under way at the Institute of Marine Sciences of the University of Miami. Establishment of this program was through the efforts of Dr. Warren J. Wisby, and to him, for
direction, advice, and encouragement, the author is especially grateful. Many other persons donated assistance and therefore deserve acknowledgment. Mr. Joseph D. Richard helped with the many technical problems, and designed much of the original instrumentation. Additional help with acoustical problems was given by Dr. John C. Steinberg, Roger Dann, John Clark, William Green, Jack Lowenstein, William Cummings, and Morton Kronengold. Sincere thanks are due the author's fellow graduate students, Samuel Gruber, Richard Aaron, Samuel Ha, and Herman Kumpf, who helped whenever needed, and to Terry Starck who provided a reliable supply of live sharks.

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**Materials**

*Experimental Animals.*—Young lemon sharks, *Negaprion brevirostris* (Poey), were used in this study. This species was chosen because of its availability, and because its behavior and size (when adult) make it potentially dangerous to man. Individual sharks used in this study ranged from two to three feet in length, and were supplied by a collector operating out of Lower Matecumbe Key, Florida.

*Apparatus.*—All of the experiments in this study, with the exception of Experiment 2(a), were conducted indoors in a plastic swimming pool, 12 feet in diameter and 3 feet deep, provided with a sand bottom. The pool was supplied with running seawater which was pumped to the laboratory building from Bear Cut, the body of water adjacent to the Institute of Marine Sciences. Experiment 2(a) was conducted outdoors in a saltwater pond measuring 50 feet long, 30 feet wide, and 5 feet deep. The bottom was composed of muddy sand.

The sound-producing equipment consisted of a Hewlett-Packard 200 CD oscillator, an Eico St40 amplifier, and a U.S. Navy J9 underwater-transmitting transducer. Sound amplitude was regulated with a precision attenuator capable of 0 to 110 dB attenuation in 1 dB steps.

The location and design of the switching equipment proved to be of great importance. Early in the study it was discovered that the sharks would respond not only to the primary signal, but also to the switching transients (clicks) and amplifier hum. Switching transients were eliminated by incorporating a transientless switch (designed by J. D. Richard) that utilized a photoconductive element. The problem with the hum was eliminated by placing all of the switching and amplitude-regulating components before the amplifier, thereby leaving the amplifier, with fixed gain, directly connected to the transducer. In this way, the hum was uniform and ever present; it formed a part of the ambient noise.
FIGURE 1. Apparatus for hearing-threshold determinations on lemon sharks, using the methods of approach conditioning and arousal conditioning, Experiments 1(a) and 1(b). A, plastic pool; B, J9 transducer; C, amplifier; D, attenuator-photoswitch unit; E, oscillator; F, hydrophone; G, sound-level meter; H, electronic filter; I, vacuum tube voltmeter; J, oscilloscope.

Sound levels were monitored with a U.S. Navy AN/PQM-1A noise-measuring set, equipped with a DT99 hydrophone. Audio waveforms were checked for distortion by inspection on a Tetronix type 502 oscilloscope. Filtered ambient-noise readings were taken, using the noise-measuring set, in conjunction with a Krohn-Hite 330-M electronic filter set for single frequencies (attenuation from center frequency was 14 to 18 dB per octave) and a Hewlett-Packard 400D vacuum-tube voltmeter. The complete arrangement, including the experimental pool, is illustrated in Figure 1.

The acoustics of the pool complicated the sound-measuring process. Since the wall of the pool, essentially an air-water interface, was a good acoustic reflector, complex interference patterns and standing waves existed. Detailed mapping of the sound amplitudes throughout the pool, at each frequency, was therefore necessary.
During the testing sessions, the shark could be found at any location within the pool. When a trial was given, therefore, it was necessary to record the shark's position at the time the sound was turned on. The true amplitude of sound at the shark's location was then determined by comparing the standard hydrophone measurement, taken during the trial, with the maps of amplitude variations for the particular frequency used.

In the experiments involving electric shock, power was supplied by a variac transformer preceded by an isolation transformer. Since it was impractical to electrify the entire pool, electrodes were harnessed directly to the shark. The harness consisted of a one-inch section of one-inch I.D. polyethylene pipe, inside of which were mounted two electrodes. The harness was loosely fitted around the caudal peduncle of the shark. The wires which were attached to the electrodes were led first to a swivel mounted on the ceiling above the pool, and then to the variac transformer.
This arrangement (Fig. 2) allowed the shark considerable freedom of movement, and a minimum of wire dragging behind.

A special arrangement was constructed for Experiment 2(a), which was conducted in the outdoor pond. The transducer was mounted on a vertical stake, at a depth of three feet. Electric shock was administered by means of a pair of electrodes mounted on either side of the transducer, at a distance of four feet apart (Fig. 3).
EXPERIMENTAL METHODS

The results of this study were obtained through the use of various conditioning techniques on free-swimming sharks. Data were gathered by direct observation of overt conditioned responses exhibited by the sharks in response to the sounds presented. The following four types of responses were used.

Approach Response (Food Reinforced).—A shark displaying this response reacted to the sound by abruptly terminating its random activities and swimming, with accelerated velocity, directly toward the transducer. The conditioning procedure was to present food (small pieces of fish) at the transducer when it was being sounded. Each shark received about four or five presentations of sound and food per day. Food was presented attached to a clip at the end of a long bamboo pole. Normally, first indications of learning came within a week, and after two or three weeks the sharks were fully trained.

When well established in a healthy, vigorous shark, the approach response was a very positive, unmistakable reaction. It was especially well suited to the orientation experiments, as the response involved swimming directly toward the sound source. A disadvantage of this response, however, was the small number of trials possible in one day, thus necessitating training and testing periods of many weeks.

Arousal Response (Food Reinforced).—This response was observed in a shark which spent much time resting quietly on the bottom, and consisted of a “start from rest” or initiation of swimming activity at the onset of the sound. First notice of this response occurred during an attempt to establish approach conditioning in one relatively sluggish shark. Although a good approach response was never achieved with this shark, the resulting arousal response appeared reliable. Whenever possible, this response was reinforced with food given either at the transducer or elsewhere.

Escape Response (Electric Shock Reinforced).—The conditioned escape response consisted of a short burst of speed (in a swimming shark) or an initiation of swimming activity (in a resting shark). The response resembled, but was less vigorous than, the unconditioned response to shock, which consisted of accelerated swimming and thrashing about, and included biting anything within reach such as the transducer, the hydrophone, or the electrode wires.

The conditioning procedure was to present the sound, wait two seconds, and then administer one or two seconds of intermittent shock. Both sound and shock were ended simultaneously. The optimum shock appeared to be about two volts across the one-inch gap between the electrodes. Much larger voltages resulted in a reaction in which the shark violently
bit the electrode wire, or its own tail, and sank to the bottom in a state of muscular tetanus.

First signs of the conditioned escape response came within two days (100 trials). Thus there was a more rapid acquisition of this response (in terms of days) as compared to the food-reinforced responses. A disadvantage, however, was that rubbing of the electrode harness caused erosion of the caudal peduncle, which often led to premature death of the shark.

**Approach-avoidance Discrimination (Food-shock Reinforced).**—The shark exhibiting this discrimination responded by approaching the transducer at the onset of a sound of one frequency, and by not approaching (avoiding) the transducer when it sounded at a different frequency. Discrimination conditioning was accomplished by pairing one frequency with food and another frequency with electric shock. The shark was given food if it arrived at the transducer during the presentation of a tone of a certain frequency (the food signal). If the shark approached the transducer during the presentation of a tone of another frequency (the shock signal), it received a mild shock from an electric field of about 15 volts across the four-foot distance between the electrodes. At the beginning of the training period, the shark, which had previously been approach conditioned, came to the transducer during both the food signal and the shock signal. The shark reacted to the shock by darting away momentarily, then quickly returning only to dart away again, and finally becoming unwilling to approach the transducer. After about one minute, however, the shark would again approach the transducer without hesitation.

First signs of reluctance to approach the transducer at the onset of the shock signal were observed after three days (15 shock trials and 29 food trials). A reliable avoidance response to the shock signal was established after six days (33 shock trials and 50 food trials). Although the shark clearly avoided the shock signal, its response to the food signal remained as good as ever.

**Experiments and Results**

**Thresholds of Hearing.**—Hearing-threshold curves were obtained from three different sharks, using a different testing procedure, as described below, for each individual shark.

**Experiment 1(a):** Using an approach-conditioned shark, thresholds were determined at one-octave frequency intervals ranging from a low of 10 cps to a high of 640 cps. Near the upper-frequency limit, half-octave intervals were used.

The procedure for threshold determination involved presenting a sound of a given frequency and amplitude, and noting whether a response occurred. If a response occurred, the next trial was given at a lower amplitude. Sound levels were reduced by approximately 5 dB per trial. This
value could be only approximate, because the shark was continually swimming about the tank in a rather random fashion, and therefore exact sound amplitudes, at the shark's location, could not be quickly determined. When a level was achieved where no response occurred, the amplitude was increased for the next trial. When an approximate threshold was obtained, several approaches to it were made. The lowest amplitude at which an unmistakable response occurred was considered the threshold value for that session.

In order to maintain a good state of conditioning, the shark was given food reward at the transducer after each positive response. A daily testing session was terminated when the shark became satiated and no longer responded vigorously. Normally this happened after about six positive responses. One day's testing, therefore, usually yielded only a single threshold at a single frequency. A total of three threshold values at each frequency were obtained using this method.

**Experiment 1(b):** Using the arousal response, thresholds were determined for a shark at the same frequencies used in Experiment 1(a). Thresholds were obtained by beginning with a sound below threshold and increasing its amplitude, in 5 dB steps, until a response was given. The lowest level at which a response occurred was considered a threshold value. Several such thresholds were obtained, at a given frequency, during each daily testing session. Approximately 12 threshold values at each frequency were obtained using this method.

Care had to be exercised in distinguishing true responses from false responses, i.e., those due to factors other than the sound. For instance, if a "start from rest" was immediately followed by rolling and scratching behavior, it was discounted as a response, because the shark may have initiated the activity as a response to a desire to scratch and not as a response to the sound. Apparent responses were also discounted if, during the intertrial intervals, the shark behaved restlessly and exhibited frequent "starts from rest" spontaneously. Undoubtedly a few false responses were included, and for this reason the mean of the individual threshold determinations was used, instead of the lowest threshold determination, as the best indication of the true threshold for this shark.

**Experiment 1(c):** Using an escape-conditioned shark, thresholds were determined, as in Experiment 1(b), by starting below threshold and increasing the amplitude, in 5 dB steps, until a response was observed. Although escape responses could usually be detected in the shark while it was swimming, trials were given only when the shark was resting on the bottom, as this allowed the more objective "start-from-rest" criterion for response. Again, it was necessary to guard against false responses, such as those caused by restlessness of the shark. Approximately 15 threshold values at each frequency were obtained by this method.
FIGURE 4. Mean hearing thresholds for the lemon shark, *Negaprion brevirostris*, determined by approach, arousal, and escape conditioning, Experiments 1(a), 1(b), and 1(c). A, expressed in absolute units; and B, relative to filtered ambient noise. Small points represent mean thresholds for each individual shark.

**Combined Results**: The best estimate of the true hearing-threshold curve of the lemon shark is given in Figure 4. This represents the mean of the mean threshold determinations made on three different sharks by the three different methods described above. The results are in general
agreement on several important points. In all three sharks the upper-frequency limit was at about 640 cps (no responses obtained one-half octave higher). The lowest threshold relative to ambient noise was always at 40 cps (mean of $-4 \text{ dB}_{\mu}b$), where it was approximately equal to filtered ambient noise and about 12 dB above spectrum-level ambient noise. The lowest threshold in absolute terms occurred at 320 cps in two sharks, and at 160 cps in another, giving a mean value at 320 cps of $-13 \text{ dB}_{\mu}b$ (14 dB above filtered ambient noise, 35 dB above spectrum-level ambient noise).

Although the lowest frequency on the threshold curve is 10 cps, this probably does not represent a lower-frequency limit of hearing. Acoustic conditions in the pool were such that, as the frequency was lowered below about 40 cps, sound was produced with diminishing efficiency. At 10 cps, very little sound energy could be produced in the pool. Even under ideal conditions for low-frequency sound production, the J9 transducer would be at reduced efficiency when operated below its normal frequency range of 40 to 20,000 cps. Conditions for sound production in the pool above 640 cps, however, were good.

The upper-frequency limit of 640 cps means that no responses were obtained at higher frequencies at the amplitudes attainable with the apparatus. If much higher amplitudes were available, then a higher upper-frequency limit might have been obtained. However, since the threshold at 640 cps is already far above (65 dB) filtered ambient noise, it seems that any ability to hear above this frequency would be of limited use to the shark. The most useful part of the shark's hearing spectrum is probably below 320 cps.

**Frequency Discrimination.**—Limits of frequency discrimination were obtained from two different sharks, using the two different methods described below.

**Experiment 2(a):** Approach-avoidance conditioning was established in the same shark that had been used in Experiment 1(a). The frequency of the food signal was 40 cps, and the initial frequency of the shock signal was 160 cps.

A convenient measure of the shark's response was the time interval between the onset of the sound and the arrival of the shark at the transducer. An arbitrary limit of 30 seconds was adopted. If the shark had not contacted the transducer within 30 seconds, the trial was ended and an arrival time of 30 seconds was recorded. Each trial was begun with the shark at approximately the same position, i.e., circling the transducer about three feet away. At full speed, the shark could cover this distance in as little as one second.

The testing procedure was to present the food and shock trials in randomized order, with intertrial intervals of roughly one minute, and to
record arrival times. Sound amplitudes were also randomly varied over a 20-dB range centered at levels chosen to have equivalent amplitudes to the shark, i.e., levels equidistant from thresholds. Thus, the shark did not receive amplitude clues and had to discriminate between the food and shock signals by frequency alone. Food and shock reinforcements were given, at the appropriate times, throughout the experiment.

Limits of frequency discrimination were obtained by reducing the difference in frequency between the food and the shock signals until no significant difference in arrival times could be observed. The initial difference of two octaves was reduced daily by lowering the frequency of the shock signal. A daily testing session lasted through about 10 presentations of food.

The results, shown in Figure 5, give the mean arrival times for each frequency of shock signal, as compared to that for the food signal for the same testing session. These means are based on approximately 20 shock and 20 food trials at each frequency. The difference in mean arrival times for the two-octave trials (160 vs. 40 cps) yielded a value of $t$ significant beyond the 99.9 per cent confidence level. The one-octave

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**Figure 5.** Frequency discrimination in a lemon shark, *Negaprion brevirostris*, determined by approach-avoidance conditioning, Experiment 2(a). Upper curve: mean arrival times for the shock signal (various indicated frequencies). Lower curve: mean arrival times for the food signal (40 cps).
Frequency discrimination in a lemon shark, *Negaprion brevirostris*, determined by escape conditioning, Experiment 2(b). Upper curve: per cent positive responses for the shock signal (40 cps). Lower curve: per cent positive responses for the neutral signal (various indicated frequencies).

(80 vs. 40 cps) and three-quarter octave (70 vs. 40 cps) trials gave differences significant at about the 97 per cent level. At one-half octave (60 vs. 40 cps) significance dropped to 92 per cent, and at one-quarter octave (50 vs. 40 cps) the mean arrival times for both signals were equal. Thus, it appears that, under the conditions of Experiment 2(a), the limit of frequency discrimination was approximately one-half octave.

**EXPERIMENT 2(b):** Utilizing escape-conditioning with a previously untrained shark, discrimination training was achieved by pairing electric shock with a 40 cps tone (the shock signal), but giving no reinforcement at tones other than 40 cps (the neutral signal). During initial training, the neutral signal was 160 cps. First signs of learning occurred after two days (25 shock trials and 100 neutral trials), and after three days (60 shock trials and 170 neutral trials) the shark was ready for testing.

The testing procedure was to present shock trials and neutral trials in random order, with intertrial intervals of one minute or more, and to
FIGURE 7. Apparatus for the determination of acoustic orientation in a lemon shark, using the method of approach conditioning, Experiments 3(a) and 3(b). A, plastic pool; B, opaque plastic curtain; C, partitions (made of wooden dowels); D, J9 transducers; E, signal-generating equipment; F, hydrophone; G, signal-monitoring equipment; H, shark in “begin-trial” position; I, transducer-partition angle.

record a response when it occurred. Again, sound amplitudes were randomly varied. Normally, more neutral trials were given than shock trials, because of the ill effects of repeated electric shock. Starting with 160 cps, the frequency of the neutral signal was reduced one step per day until the percentages of responses for the two signals were statistically indistinguishable.

The results of this experiment are presented in Figure 6, and are based on approximately 40 shock trials and 70 neutral trials at each frequency. At the two-octave level (160 vs. 40 cps) the shark exhibited excellent discrimination, responding to the shock signal in 91 per cent of the trials but to the neutral signal only 6 per cent of the time. This difference gives an extremely significant chi-square value. The difference at one octave (80 vs. 40 cps) is less, 77 per cent for the shock signal and 14 per cent for the neutral signal, but still significant beyond the 99.9 per cent level.
The smallest difference tested, one-half octave (60 vs. 40 cps), resulted in 89 per cent responses for the shock signal and 63 per cent responses for the neutral signal, with significance dropping to 99.2 per cent. Unfortunately, testing had to be discontinued before smaller differences could be tried, because of deterioration of the shark's health. From extrapolation of the data, however, it appears that the shark's limit of discrimination under the conditions of this experiment lay somewhat below one-half octave, probably between one-half and one-quarter octave. This is in general agreement with the results of Experiment 2(a).

Acoustic Orientation.—Early in the study it was noted that the approach-conditioned sharks appeared able to localize the source of the sound, thus enabling them to swim directly toward the transducer. Since these observations did not rule out the possibility of visual orientation, the following experiments were designed to determine if directional hearing did exist and, if so, to what degree of accuracy.

For these experiments, the pool was modified by including an opaque plastic curtain, behind which two J9 transducers were placed so as not to be visible to the shark. This curtain, suspended from a float and weighted at the bottom, formed a "false wall" about one foot inside the real wall of the pool. In addition, two partitions were included, one of which served to divide the far side of the pool into right and left halves, and the other of which served to guide the shark into a position suitable for the beginning of each trial. These partitions were constructed of wooden dowels which were vertically mounted and spaced about two inches apart. The entire arrangement, including hydrophone and transducers, is shown in Figure 7.

The shark used in these experiments was the same well-trained individual used in Experiments 1(a) and 2(a). This animal adapted quickly to the new apparatus in the pool and responded, as before, by accelerated swimming toward the sound source, even though the transducer was now hidden behind the plastic curtain. Food reward was given to the shark when it arrived at the curtain near the transducer.

Experiment 3(a): The design of this experiment allowed the shark two possible choices, correct or incorrect. A correct response occurred if the shark arrived initially at the curtain on the side of the partition having the active transducer. A trial was begun only when the shark was in the "begin trial" position (Fig. 7), where it was in line with the partition and equidistant from both transducers. At this point the shark was approximately seven feet from either transducer. Choice of active transducer, for a particular trial, was determined randomly.

During the experiment, the transducers were located at various angles from the partition (Fig. 7). At an angle of 45 degrees, for instance, one transducer was 45 degrees to the right of the partition and the other was
Figure 8. Acoustic orientation (per cent incorrect responses) in a lemon shark, *Negaprion brevirostris*, at various transducer-partition angles, Experiment 3(a). Mean orientation error is found at the 25 per cent level for incorrect response.

45 degrees to the left. Thus, if the shark's approach error was more than 45 degrees, there was a 50 per cent chance that it would arrive at the curtain on the incorrect side of the partition. There was also a 50 per cent chance, however, that the shark would miss the transducer by more than 45 degrees on the other side, a circumstance still resulting in a correct response. Furthermore, if the shark's mean error was 45 degrees, then one would expect 25 per cent incorrect responses. Therefore, the shark's mean approach error, in degrees, was equal to that transducer-partition angle which resulted in 25 per cent incorrect responses.

Orientation determinations were conducted at three separate frequencies: 40 cps, 80 cps, and 320 cps. These represented the low, middle, and high parts of the shark's hearing spectrum. Original plans to use 20 cps as the low frequency had to be abandoned, because the shark could not detect 20 cps well enough in the pool at the distances involved.

Results, shown in Figure 8, were based on 52 individual trials at each frequency, and indicated that the shark possessed good orientation ability, being able to swim toward the source of sound with a mean error of
TABLE 1

COMPARISON OF MEAN ERRORS OF ORIENTATION TO SOUNDS FOR A LEMON SHARK, *Negaprion brevirostris*, AT SEVERAL FREQUENCIES

<table>
<thead>
<tr>
<th>Frequency (cps)</th>
<th>Mean orientation error (degrees)</th>
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<tbody>
<tr>
<td></td>
<td>Experiment 3(a)</td>
</tr>
<tr>
<td>40</td>
<td>7.0</td>
</tr>
<tr>
<td>80</td>
<td>7.5</td>
</tr>
<tr>
<td>320</td>
<td>14.0</td>
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<tr>
<td>Combined frequencies</td>
<td>9.5</td>
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about 9.5 degrees. The shark appeared able to orient somewhat better at 40 and 80 cps than at 320 cps. A test of the difference between 320 cps and either 40 or 80 cps, at the angles where incorrect responses were made (22.5, 10, and 5 degrees), gave a chi-square value significant at the 93 per cent level of confidence.

**Experiment 3(b):** In this experiment, the observer directly estimated the shark's error in swimming toward the hidden source of sound. The parameter estimated was the angle of error of the initial approach direction. Upon hearing the sound, the shark's usual behavior was to turn and swim in a straight line approximately toward the source. If the shark was not directly headed toward the transducer, it would make corrections as it neared the curtain. Normally, however, the first half of the shark's path was straight, and it was this initial approach direction that was compared to the actual direction of the transducer.

The testing procedure was to play a sound, estimate the shark's error in approach, and then reward the shark upon its arrival at the curtain near the transducer. After every two trials, the location of the transducers behind the curtain was changed. Again, a trial was not started until the shark was in the "begin trial" position.

The results of this experiment, which were based on 12 trials at each frequency, agreed well with the results of the previous experiment. Table 1 gives the results of this experiment, and compares them with the results of Experiment 3(a). The mean orientation error, combining all frequencies, was calculated to be 7.1 degrees. Again, it appeared that orientation to the lower frequencies was slightly more accurate than orientation to 320 cps. A *t* test of the mean error at 40 cps and 320 cps showed a difference significant at the 96 per cent confidence level. No significant difference was shown between 80 cps and 320 cps or between 40 cps and 80 cps.
DISCUSSION

Thresholds of Hearing.—The hearing-threshold curve (Fig. 4) obtained in this study is, thus far, the only such curve available for the lemon shark. It differs in several respects from threshold curves reported for other species of sharks.

Kritzler & Wood’s (1961) audiogram for the bull shark was produced using an approach response similar to that used in Experiment 1(a) of this study. Their results show maximum sensitivity relative to ambient noise in the band of 400 to 600 cps. Although an exact comparison to the results of this study cannot be made because of differences in acoustic environments, it is evident that this value is much higher than the 40 cps for the lemon shark. Their upper-frequency limit is also higher than the 640 cps value obtained in this study. Their low-frequency limit of 100 cps may have been influenced by the poor low-frequency characteristics of their underwater speaker, but it probably indicates much less sensitivity than in the lemon shark. In the areas of greatest sensitivity, however, both threshold curves approximate filtered ambient noise.

The hearing range reported by Olla (1962) for small hammerhead sharks in an 18-foot-diameter plastic pool more nearly coincides with that found for the lemon shark. He found an upper-frequency limit of 750 cps, which approximates that found in this study. Although no absolute threshold values were given, the maximum sensitivity occurred at 250 to 375 cps, apparently the lowest frequencies used. This compares favorably with the 320 cps “absolute” sensitivity peak for the lemon shark.

The preliminary results of Davies et al. (1963) are drastically different from those of Kritzler & Wood (1961), Olla (1962), and the present study. The curve reported by Davies et al. extends from 50 cps to 7,000 cps, with slightly greater sensitivity at 50 cps, presumably the lowest frequency tested. At 50 cps, the threshold value approximates filtered ambient noise. The high-frequency end of the curve, however, remains near ambient noise and from 3,000 to 7,000 cps is perfectly flat. This condition is highly unlikely for a hearing-threshold curve. Moreover, the octave-band noise levels shown do not decrease with increasing frequency as is normal. Finally, the curve is based on the collective responses of a group of 25 to 30 sharks of four different species. Until further evidence is available, or more experimental details are given, the results of Davies et al. must be regarded as questionable.

The most carefully produced audiogram available for any fish is probably that produced by Tavolga & Wodinsky (1965), using avoidance conditioning with the blue-striped grunt, Haemulon sciurus. Maximum sensitivity was found at 50 to 200 cps (-20 dBµb), the lowest frequencies tested, and the upper-frequency limit was 1,000 cps. Considering the lower ambient-noise levels and lack of data below 50 cps, their threshold
curve does not appear very different from that for the lemon shark. Again, however, exact comparison is impossible because of the difference in acoustics between their five-gallon, sound-insulated aquarium, and the 2,500-gallon plastic pool used in this study. In an earlier report (Tavolga & Wodinsky, 1963), threshold curves were given for nine species of marine teleosts which showed maximum sensitivities in the range of 300 to 800 cps, with upper-frequency limits of 1,000 to 1,200 cps (one at 2,800 cps).

An important matter, which has been largely neglected in the past, is that of the masking effect of background noise. Since the aquatic environment is normally one of considerable ambient noise, the ability of a fish to detect a sound probably depends more on its ability to discriminate a signal from noise than on its ability to detect low absolute amplitudes of signal. Thus, threshold determinations are greatly affected by the level of background noise, and comparisons of hearing sensitivities of different species are relatively meaningless unless thresholds were taken at equivalent levels of background noise.

Ambient noise levels in Tavolga & Wodinsky's (1963) sound-insulated aquarium were reported as much as 35 dB lower than in Kritzler & Wood's (1961) shark pens or in the pool used in this study. Tavolga & Wodinsky were able, therefore, to obtain thresholds with less interference from background noise. Their lowest threshold (for *Eqetus*, -40 dBpL at 600 cps) was still about 5 dB above filtered ambient noise, and for most of the species tested, thresholds were well above ambient noise. Further work by Tavolga (1966) emphasizes the effect of masking noise on hearing thresholds in fish. Absolute thresholds of hearing for the fresh-water catfish, *Amiurus (=Ictalurus) nebulosus* (Poggendorf, 1952), have been reported as low as -50 dBpL. If this value is correct, then ambient noise levels, which were not given, must have been exceedingly low.

Whether the thresholds for the lemon and bull sharks would become lower at lower ambient noise is an interesting question. Nelson's (1965) experiments with unconditioned cardiac responses to sounds in the lemon shark indicated the importance of signal-to-noise ratios, at least at relatively high levels of noise. An absolute threshold of hearing, however, cannot be lowered by any degree of background quieting, and whether this level has been reached for the lemon and bull shark can be determined only by further experimentation at lower noise levels.

An understanding of the relationship between acoustic pressure and displacement in the water medium surrounding a sound source is essential to an understanding of the mechanisms of hearing in fish. Harris & van Bergeijk (1962) and Harris (1964) have shown that the near field of a sound source is characterized by displacements of much greater magnitude than one finds in the far field at the same acoustic pressure. Furthermore,
since pressure-displacement relationships within the near field vary with the size and shape of the container, displacement can be conveniently calculated from pressure only in the far field.

Ideally, sound amplitude measurements should be made with both pressure-sensitive and displacement-sensitive hydrophones. Suitable displacement hydrophones, however, have only recently been developed (Liebermann & Rasmussen, 1964) and are not yet readily available. In past studies of hearing in fish, all measurements of sound amplitudes were made only with pressure-sensitive hydrophones.

It has been postulated that the swim bladder of teleost fishes acts as a receiver of acoustic pressure (Harris & van Bergeijk, 1962; Alexander, 1966). The most plausible mechanism for this is that, under the influence of acoustic pressure fluctuations, the swim bladder experiences volume fluctuations. These, in turn, cause in-and-out movements of the bladder wall which are propagated, as displacements, to the hearing organs of the fish. Thus, the swim bladder can be regarded as a pressure-to-displacement converter, responsible only for the initial reception of pressure. The final means of stimulation of any mechanoreceptor end organ, it must be remembered, is always mechanical deformation (displacement).

In fishes lacking swim bladders, i.e., all elasmobranchs and a few teleosts, the above-postulated mechanism of pressure reception would not apply. The most likely mechanism of hearing in these fishes would be detection of the sound as displacements arriving directly from the source. The possibility of some unknown mechanism of pressure-displacement conversion in these fishes, however, should not yet be ruled out.

If hearing in fish involves initial reception of the sound as displacements, either wholly or in part, and there is some experimental evidence for this (Harris & van Bergeijk, 1962; Banner, 1966; and Enger, 1966), then amplitude thresholds based on pressure measurements in the near field may have to be regarded as less meaningful. Also open to question would be the validity of comparisons of hearing sensitivity of different species if the thresholds were determined under different sets of pressure-displacement conditions. Thus, until the pressure-displacement question is more fully explored, it seems best to regard existing amplitude thresholds, including those of this study, as only provisional indications of true hearing sensitivity.

Frequency Discrimination.—In testing frequency discrimination, care must be taken to insure that the two signals are discriminated by frequency and not by amplitude. Thus, while frequencies are changed, amplitudes must be kept constant. The amplitude to consider, however, is not absolute amplitude measured in physical terms, but subjective amplitude or loudness. In infrahuman animals, this can be done by setting the amplitude of each frequency at some constant number of decibels above threshold.
Unless the hearing-threshold curve is precisely known, however, it is difficult to arrive at accurate equivalent loudnesses. Even if the subjective loudness of the two signals differs by only a small amount, it is possible that this small amount is being discriminated. Thus, supposed frequency-discrimination limits may actually be amplitude discrimination limits, especially since amplitude differences usually diminish along with frequency differences.

In the present study, this difficulty was overcome by varying the amplitudes of both signals, in random fashion, about central values chosen to be equivalent in loudness. Kleerekoper & Chagnon (1954), who obtained limits of frequency discrimination for the creek chub, *Semotilus atromaculatus*, stated that their amplitudes were kept constant, but did not say whether this meant absolute or subjective amplitudes. They did, however, perform other experiments (no details given) to ascertain that the fish responded to frequency rather than to amplitude. Wohlfahrt (1939), using the minnow, *Phoxinus laevis*, was convinced that his fish distinguished frequency and not amplitude, because of additional experiments in which he could not obtain discrimination by amplitude alone. Dijkgraaf & Verheijen (1949), however, showed that *Phoxinus* was capable of amplitude discrimination as well as frequency discrimination.

Results of frequency-discrimination determinations depend greatly on the temporal spacing of the two signals to be distinguished. Best discrimination is obtained when the two signals are very close together. Wohlfahrt's (1939) and Dijkgraaf & Verheijen's (1949) method, of using a pulsed sound of a single frequency for one signal and a pulsed sound of that frequency alternated with a different frequency for the other signal, presented very close temporal spacing. For *Phoxinus*, they obtained limits of frequency discrimination of 0.08 octave, and 0.04 octave, respectively. Kleerekoper & Chagnon (1954) used a time interval of about eight to ten seconds, and obtained a limit of 0.2 octave for *Semotilus*. When they used longer intervals (no times given) they obtained an "absolute" frequency-discrimination limit of 0.4 octave. This value compares well with the limit of about 0.5 octave for the lemon shark (time interval about one minute) as, in both cases, the animal had to "remember" frequencies for considerable lengths of time.

After consideration of the anatomy of the inner ear of sharks, Davies *et al.* (1963) concluded that "it seems most unlikely that sharks are able to discriminate frequencies or pitch as they apparently possess no organ equivalent to the basilar membrane, which is responsible for frequency discrimination in the human." Such reasoning would deny frequency discrimination in teleosts, as well as in sharks, and must be considered invalid. Davies *et al.* also claimed behavioral support for their conclusion, but just what data this was based on is not clear.
Although the presence of frequency discrimination in fish appears to be well established, nothing is known of the mechanism. It is most probably not due to stimulation of a "place" on the receptor organ, as on the basilar membrane in the cochlea of mammals, but rather a function of the rate of displacement oscillation of the receptor organ.

**Acoustic Orientation.**—There are several methods by which animals can reach the source of a sound. The simplest is gradient seeking, in which the animal samples amplitudes, in trial and error fashion, until the source is located. Gradient seeking is possible in animals possessing just one nondirectional receptor, and is characterized by a very erratic path toward the source.

A better way of locating a sound source is by means of directional hearing, a process in which the animal senses the direction of the source. The animal is thus able to travel straight toward the source. In most terrestrial vertebrates, directional hearing is accomplished with two nondirectional receptors. The essential factor is a detectable difference in arrival times or in amplitudes at the two receptors. In humans, a sound is localized because the nearer ear receives the sound first and at a higher amplitude (the other ear is both farther away and in a sound shadow created by the head). This mechanism is theoretically less advantageous underwater, however, because of the greater velocity of sound and because similarities of acoustic properties of flesh and water tend to minimize amplitude differences between the ears (less sound shadow).

Van Bergeijk (1964) discusses the theoretical mechanisms of directional hearing. He points out that a two-receptor array (of nondirectional receptors), operating on time-of-arrival differences or amplitude differences, can locate the source angle, but usually with a left-right or front-back ambiguity. To locate unequivocally the source, the two-receptor array must be movable (rotatable) so that a second source angle can be obtained with which to compare the first. If the array consists of three or more receptors, the source can be unequivocally localized without moving the array. Sharks possess more than three receptors, although the number used in localization is not known, and sharks characteristically swim with a left-right scanning motion of the head.

Pressure is not a vector quantity, and a single pressure receptor, therefore, cannot be directionally sensitive. In teleost fishes, in which the swim bladder is believed to act as a single pressure-to-displacement converter, it has been theorized that directional hearing is not possible using pressure reception. Past studies under far-field conditions by von Firsch & Dijkgraaf (1935) and Reinhardt (1935) substantiate this theory. Kleerekoper & Chagnon (1954) concluded that the creek chub could locate the source of sound. Their experiments, however, were conducted well into the near field and, furthermore, the responses described were probably due to gra-
dient seeking rather than to directional hearing. Certain responses suggesting directional hearing in squirrelfish, *Holocentrus rufus*, and toadfish, *Opsanus tau*, have been observed by Winn et al. (1964), but it is not clear if these were valid responses or if true far-field conditions were involved. Thus, if far-field directional hearing exists in teleosts, it has yet to be demonstrated.

Displacement is a vector quantity and would be capable of imparting directional information to a single receptor if a suitable one existed. Such a directional receptor may not be necessary, however, because fish possess numerous nondirectional displacement receptors of several types including the inner ears, the lateral-line organs, and the general tactile sensors. A fish, therefore, has more than the two or three receptors necessary for localization, especially well into the near field where displacements are great enough to stimulate many of these receptors. When displacements approach far-field magnitudes, however, fewer receptors may be available with sufficient sensitivity, although the most sensitive receptors, presumably the inner ears, would operate at the lowest detectable sound level.

The involvement of the swim bladder suggests a possible mechanism to explain the observed differences in far-field directional hearing capabilities of teleosts and sharks. In teleosts, the dominant displacements reaching the receptor organs are those produced when the swim bladder pulsates (changes volume) in response to acoustic pressure. Thus, all the receptor organs and the swim bladder can be taken collectively as one nondirectional sound receptor. In sharks, which do not possess swim bladders, the displacements reaching the receptor organs are those coming directly from the source, and arriving at the near receptors earlier than at the far receptors. A small amount of shadowing may also occur, due to the acoustic properties of the shark's body. According to this theory, teleosts would be capable of directional hearing only when the displacements reaching them directly from the source are large enough to outweigh those displacements coming from the swim bladder. This would probably occur only well within the near field.

In theory, directional hearing could be based on receptors which are, in themselves, sensitive to displacement direction. In such a case, a single receptor would be capable of localizing a source, but with a 180 degree ambiguity, whereas a two-receptor array would be necessary for unequivocal localization. Proof of the existence of directional hearing based on directional receptors, however, is lacking, although some individual hair-cell receptors are morphologically polarized in a way suggesting directional sensitivity (Flock, 1964; Flock & Duvall, 1965). Davies et al. (1963) suggested that, in certain sharks, because the plane of orientation of the sensory epithelium of the right sacculus is different from the left, displacements in a given direction may stimulate one sacculus more than
the other. If this theory is correct, both sacculi may be taken collectively as a single directional receptor, because the signal parameter utilized would be displacement direction (measured essentially at a single point), rather than time-of-arrival or amplitude differences (which must be measured at two points separated by some considerable distance).

Localization of an underwater sound source by displacement direction involves further complications (van Bergeijk, 1964). All far-field displacements and near-field displacements from a monopole source (changing volume, fixed center of gravity) are in the direction of the source, i.e., the displacements are in a radial direction. Thus, measurement of the displacement direction will yield information about the location of the source. The J9 transducer used in this study, however, more nearly resembles a dipole source (fixed volume, changing center of gravity) which yields radial displacements only in the axis of vibration of the source. At 90 degrees to the axis of vibration, the displacements are parallel to the axis, which is 90 degrees from a radial direction. At other locations the displacement direction is determined by both radial and angular components. Thus, measurement of displacement direction at one point in the near field of an unknown type of sound source yields no definite information about its location. Because of these objections, it seems unlikely that localization in sharks is based on detection of displacement direction, but rather on detection of time-of-arrival or amplitude differences.

The shark in Experiment 3 was able to locate the hidden J9 transducer with considerable accuracy within the near field. The axis of the transducer, however, was always in line with the shark when the sound was turned on. Thus, the directive responses always occurred in the axis where displacements were radial. Whether the shark could localize the transducer when not in its axis was not determined. The shark appeared able to do so in Experiment 1(a), but it can be argued that the directionality of these responses could have been based on visual cues, since the transducer was not hidden. Proof of ability to determine the direction of a dipole source in the near field, regardless of its position, would be evidence in favor of localization by time-of-arrival or amplitude differences.

From the descriptions given, the orientation responses obtained by Kritzler & Wood (1961) and Davies et al. (1963) appear similar to those observed in this study. In both cases, distances were great enough to include far-field conditions at some of the frequencies used. Kritzler & Wood’s three identical sources at three different positions served to eliminate the possibility that the shark located the active transducer by vision alone (Wood, personal communication). Davies et al., however, did not report having such a control. The directional responses from distances of up to 200 yards, observed by Wisby & Nelson (1964), occurred well into the far field and well beyond the limit of visibility. Thus, evidence
from the present study and from those mentioned above conclusively demonstrates directional hearing in sharks in both the near and far fields.

Considering all aspects of the problem, it seems most likely that the directional-hearing mechanism in sharks involves detection of the signal (probably as direct displacements, but possibly as pressure converted to local displacements by some unknown means) by two or more non-directional displacement receptors, with the shark being capable of utilizing time-of-arrival or amplitude differences between these receptors.

**SUMARIO**

**UMBRALES AUDITIVOS, DISCRIMINACION DE FRECUENCIA Y ORIENTACION ACUSTICA EN EL TIBURON GALANO,** *Negaprion brevirostris* (Poey)

Se condujeron experimentos en ejemplares jóvenes de tiburón galano, *Negaprion brevirostris* (Poey), con el propósito de determinar sus umbrales de audición, habilidad para discriminar frecuencias y su habilidad para orientarse hacia la fuente del sonido. Se obtuvieron datos por observación directa de los siguientes tipos de respuestas condicionadas al sonido.

Las respuesta de acercamiento (reforzada con alimentos) consistió en que un tiburón terminó abruptamente sus actividades al azar y nadó con velocidad acelerada, directamente hacia la fuente del sonido. La respuesta de excitación (reforzada con alimentos) consistió en un “inicio a partir del descanso” o inicio de actividad natatoria en un tiburón que previamente se encontraba descansando. La respuesta de huida (reforzada eléctricamente) consistió en un pequeño aumento brusco de velocidad en un tiburón que estaba nadando, o en un inicio de actividad natatoria en un tiburón que estaba previamente descansando. En la respuesta de acercamiento-evasión (reforzada con alimentos y electricidad) el tiburón respondió acercándose a la fuente de sonido cuanto éste tuvo una cierta frecuencia y no acercándose, *i.e.*, evadiendo la fuente de sonido cuando éste tuvo una frecuencia diferente.

Los umbrales de audición se obtuvieron de tres tiburones diferentes, usando condiciones de acercamiento en el primero, condiciones de excitación en el segundo y condiciones de huida en el tercero. La media de la curva del umbral de audición se extendió desde 10 cps (o menos) hasta alrededor de 640 cps, con la menor sensibilidad de audición por arriba de los 320 cps. La mayor sensibilidad auditiva relativa al ruido ambiente tuvo lugar a 40 cps donde la media del valor del umbral (−4 dBμb) se aproximaba al ruido ambiente filtrado. La audición más sensible en términos absolutos tuvo lugar a 320 cps cuando la media del valor del umbral (13 dBμb) fue alrededor de 14 dB por arriba del ruido ambiente filtrado.

La discriminación de frecuencia fue investigada en dos tiburones, usando los métodos de acercamiento-evasión condicionado y huida condicionada.
El límite de discriminación de frecuencia estuvo a, o algo por debajo, de la mitad de una octava en los límites de las frecuencias de 40 a 60 cps, cuando el tiempo de intervalo entre presentaciones fue aproximadamente un minuto.

Usando un tiburón condicionado al acercamiento en una piscina con fuentes de sonido escondidos detrás de una cortina plástica opaca, se determinó que el tiburón pudo orientarse hacia los sonidos, a distancias de alrededor de siete pies, con un valor medio de error de aproximación de alrededor de 8 grados. El tiburón lució capaz de orientarse algo mejor hacia sonidos de baja frecuencia (40 cps, 5.2 grados; 80 cps, 6.5 grados) que a sonidos de más alta frecuencia (320 cps, 13.4 grados).

**LITERATURE CITED**

**ALEXANDER, R. McN.**

**BACKUS, RICHARD H.**

**BANNER, ARNOLD**

**CLARK, EUGENIE**

**DAVIES, DAVID H., J. P. A. LOCHNER, AND E. D. SMITH**

**DIJKGRAAF, SVEN**

**DIJKGRAAF, SVEN AND F. J. VERHEIJEN**

**EIBL-EIBESFELDT, IRENAUS AND HANS HASS**

**ENGER, PER S.**
1966. The near-field effect on sound thresholds in goldfish. Conference on Lateral Line Detectors. (abstract circulated at meeting.)

**FLOCK, AKE**

**FLOCK, AKE, AND ARNOLD J. DUVALL**

**HARRIS, GERALD G.**
1964. Considerations on the physics of sound production by fishes. *In:*
NELSON: Hearing in Lemon Sharks


HARRIS, GERALD G. AND W. A. VAN BERGEIJK

HOBSON, EDMUND S.

KLEEREKOPER, H. AND E. C. CHAGNON

KRITZLER, HENRY AND LANGLEY WOOD

LIEBERMANN, L. N. AND R. A. RASMUSSEN

LIMBAUGH, CONRAD

LOWENSTEIN, OTTO

MOULTON, JAMES M.

NELSON, DONALD R.

NELSON, DONALD R. AND SAMUEL H. GRUBER

OLLA, BORI

PARKER, GEORGE H.

POGGENDORF, DIETRICH

RANDALL, JOHN E.
REINHARDT, FELIX

TAVOLGA, WILLIAM N.

TAVOLGA, WILLIAM N.

TAVOLGA, WILLIAM N. AND JEROME WODINSKY

VAN BERGELJK, WILLEM A.

VILSTRUP, THURE

VON FRISCH, KARL AND SVEN DIJKGRAAF

WINN, HOWARD E., JOSEPH A. MARSHALL, AND BRIAN HAZLETT

WISBY, WARREN J. AND DONALD R. NELSON

WISBY, WARREN J., JOSEPH D. RICHARD, DONALD R. NELSON, AND SAMUEL H. GRUBER

WOHLFAHRT, THEODOR A.

WRIGHT, BRUCE S.