REACTION OF TUNAS AND OTHER FISHES TO STIMULI

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Explanatory Note

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United States Department of the Interior, Oscar L. Chapman, Secretary Fish and Wildlife Service, Albert M. Day, Director

REACTION OF TUNA AND OTHER FISH TO STIMULI-1951

PART II: OBSERVATIONS ON THE CHEMORECEPTION OF TUNA ... by P. B. van Weel

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Note.---This report is also Contributions Nos. 22-26, of the Hawaii Marine Laboratory, University of Hawaii

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PART I

BACKGROUND, AND SUMMARY OF RESULTS_/

by

Albert L. Tester Professor of Zoology University of Hawaii

INTRODUCTION

At the instigation of O. E. Sette, Director, Pacific Oceanic Fishery Investigations, an agreement concerning a study of the reactions of tuna to stimuli was completed between the United States Department of the Interior, Fish and Wildlife Service, and the University of Hawaii, Honolulu, T. H., on January 19, 1951 (Contract No. I6fw-13331). The agreement called for "(a) the search of the literature on tuna reactions; (b) the development of methods for holding and caring for the various species in captivity; and (c) the study of reaction of the fish, individually and in groups or schools, to stimuli such as light, sound, chemicals and electricity, with emphasis on the study of stimuli and reactions which promise to have application to fishing operations."

A search of the literature failed to reveal any papers dealing directly with the reactions of tuna to stimuli. However, many references to the reactions of both freshwater and marine fish to stimuli of various kinds were obtained. These are on file at the Department of Zoology and Entomology, University of Hawaii.

Moderate success was encountered in establishing tuna and other fish in captivity. This has been dealt with in a separate publication (Tester 1952).

The papers which follow in this report deal with the completion of the third part of the contract, insofar as time and funds were available during the period of one year.

We wish to express our sincere thanks to O. E. Sette, Director, and to Dr. J. L. Kask and Dr. W. F. Royce, Pacific Oceanic Fishery Investigations, for their helpful suggestions and assistance. We are also indebted to Dr. R. W. Hiatt, Director, Hawaii Marine Laboratory, for general advice and assistance.

L/ Contribution No. 22 of the Hawaii Marine Laboratory, University of Hawaii

EXPERIMENTAL FISH

The experiments on chemical, light, and sound reactions were conducted on two species of tuna: the yellowfin or <u>ahi</u> (<u>Neothunnus</u> <u>macropterus</u>) and the little tunny (henceforth called "tunny") or <u>kawakawa</u> (<u>Euthynnus yaito</u>), which were established in captivity in a concrete tank at the Hawaii Marine Laboratory, Coconut Island, Oahu, over a 7-month period. The experiments on electrical reaction were performed on the "mountain bass" or <u>aholehole</u> (<u>Kuhlia sandvicensis</u>), for reasons which will be discussed elsewhere.

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The concrete tank in which the tuna were confined (fig. 1, reproduced from fig. 5, Tester 1952) is partially sunk in the ground; it has smooth 6-inch concrete walls and bottom; it is 34.7 feet long. 10.8 feet wide, and 3.8 feet deep at the north end and 4.0 feet deep at the south end; its volume is 10,663 gallons; the rate of flow of the saltwater supply is about 25 gallons per minute; the inlet, near the northwest corner, is directed horizontally (towards the south) at a depth of about $2\frac{1}{2}$ feet; the outlet is a notch cut in the top of the south wall at its center. Baffles consisting of 3 x 4-foot galvanized iron sheets, painted white, are placed across three corners; a larger baffle, 3 x 10 feet, also painted white is placed across the fourth or northeast corner for the purpose of housing equipment for light reaction studies. A $\frac{1}{2}$ -foot fence, with upright posts and horizontal, spaced plank bars, surrounds the tank at its upper edge. Towards the end of the summer the fence was lined with chicken wire to keep the fish from jumping through, and the top was similarly covered to keep visitors from throwing stones at the fish. Two 60watt bulbs were suspended above the tank and were lit from dusk to daybreak.

During the experimental work varying numbers of tuna (from one to seven) were present in the tank, as shown in table 1. As their reactions to stimuli were dependent on their "state of health," their history is briefly reviewed.

Yellowfin No. 1, the subject of most of the experiments, was introduced to the tank on June 20, 1951, started feeding on July 2, 1951, and was in excellent condition until about October 31, 1951. Following that date it took less and less food, and finally ceased feeding. The normally shiny, black skin became whitish and distended, as if the body were swollen. The swelling around the eyes made the eyeballs appear to sink within the sockets, and interfered with the tuna's vision. During late December and early January the yellowfin resumed a desire to feed; although it would snap at the food, it would invariably miss. It died on January 13, 1952, and in addition to being puffy and swollen, was found to have been blind in one eye. Its initial weight was estimated at about 5 pounds; its weight at death was 11 pounds.



FIG. I. CONCRETE TANK IN WHICH YELLOWFIN AND TUNNY WERE CONFINED.

15/1/6 - 15/1/6/9 5/1/2/9 - 15/05/9 5/1/2/9 - 15/05/9 5/1/2/9 - 15/05/9

6/20/51 = 8/22/51 1 = - 8/23/51 = 8/27/51 2 = - 8/28/51 = 8/29/51 2 1 8/30/51 = 9/2/51 2 4 9/3/51 = 9/4/51 2 5 9/5/51 = 11/11/51 1 5	
מ מ מ ת	Yellowfin No. 2 started feeding 8/26/51 Tunny No. 1 started feeding 9/1/51 Tunny No. 2, 3, and 4 started feeding 9/1/51 Tunny No. 5 started feeding 9/3/51 Yellowfin No. 2 jumped from tank 9/4/51 all other fish feeding and in excellent condition to 10/31/51
N N N H	<pre>Tunny No. 1 started feeding 9/1/51 Tunny No. 2, 3, and 4 started feeding 9/1/51 Tunny No. 5 started feeding 9/3/51 Yellowfin No. 2 jumped from tank 9/4/51; all other fish feeding and in excellent condition to 10/31/51</pre>
ର ର ମ	<pre>Tunny No. 2, 3, and 4 started feeding 9/1/51 Tunny No. 5 started feeding 9/3/51 Yellowfin No. 2 jumped from tank 9/4/51; all other fish feeding and in excellent condition to 10/31/51</pre>
<u>ମ</u> ନ	Tunny No. 5 started feeding 9/3/51 Yellowfin No. 2 jumped from tank 9/4/51; all other fish feeding and in excellent condition to 10/31/51
I	Yellowfin No. 2 jumped from tank 9/4/51; all other fish feeding and in excellent condition to 10/31/51
11/12/51 = 12/6/51 1 2	Three tunny died; other tunny and yellowfin No. 1 listless and not feeding well
12/7/51 = 12/8/51 1 1 1	Another tunny died; yellowfin No. 1 apparent- ly hungry and attempting to feed
12/9/51 = 1/13/52 1 =	Remaining tunny diedş yellowfin No. 1 apparently hungry but cannot focus on food died 1/13/52
,	

Yellowfin No. 2 was introduced on August 22, 1951, started feeding 3 days later, but jumped from the tank and died on September 4, 1951. It was in excellent condition during its short life in captivity. Its estimated weight was about 6 pounds.

The five tunny were introduced between August 27, 1951 and September 2, 1951, and started feeding within 1 to 3 days. They were in excellent condition until the end of October, following which they gradually became listless and fed only occasionally. Three died on or about November 12, one on December 6, and the last one on December 8, 1951. During their moribund condition they lost their bright coloration but otherwise (apart from their behavior) seemed normal in appearance. The reason for the sickness and subsequent mortality is unknown; it followed a period of cold, wet weather. During their period of confinement, the tunny increased from an initial weight of about 2 pounds to a final weight of 3 to 4 pounds.

Except prior to experiments on chemoreception, the tuna were fed regularly once a day. Notes on their feeding and schooling behavior are included in the reports which follow.

It was planned to study the reaction of tuna in large Pond No. 5 (Tester 1952) to light and sound stimuli during January 1952, using one yellowfin and one tunny. These were the survivors of a mortality which occurred, as in the concrete tank, during November. The survivors were feeding, and were apparently in good condition during December and the first part of January. A few experiments with sound were performed but, unfortunately, before the series could be completed, the tunny disappeared (about January 22, 1952) and the yellowfin died (about January 30, 1952), again following a period of cold, wet weather. No experiments with light were conducted in Fond No. 5.

SUMMARY OF RESULTS

Dr. P. B. van Weel, studying chemorecption in tuna, found that both the yellowfin and tunny have a well-developed sense of small or taste whereby they may be attracted to certain food substances. They were strongly attracted to clear, colorless extracts of tuna flesh. Moreover, it was found that the attractant was contained in the "protein" rather than in the "fat" fraction of the clear extract. In general, the reactions of the tunny were more pronounced than those of the yellowfin. On the other hand, there was no positive reaction of either species to "conditioned" water in which baitfish had been living, nor to extracts of either baitfish or squid. Two chemicals, other than food substances, were tried -- asparagine, a possible attractant, and copper acetate, a known shark repellent. The former did not prove to be an attractant. The latter was a repellent to tuna, although its effect was not as pronounced as on fish of other species which were also present in the tank. Future research should be directed at identifying the particular "protein" substance in the clear extract of tune flesh which acts as the attractant. If this can be isolated and prepared in large quantities, it could be used in attempting to attract tune to the stern of a fishing boat at sea.

Dr. Sidney C. Hsiac studied the reaction of the tuna be artificial light generated from an arc lamp, a projection lantern, and electric light bulbs. His experiments were performed after dark, with the tank illuminated constantly by two 60-watt bulbs. He found that both yellowfin and tunny were attracted to continuous white light over a range of moderate intensity (about 70 to 450 foot candles). However, they were not attracted by a light of weaker intensity, and they were repelled by a light of stronger intensity. Both species were attracted to colored lights of moderate intensity, but to no greater extent than to white light. Similar results were obtained with interrupted white light. There appeared to be no relationship between the strength of the reaction and the frequency of interruption of the light. It was noted that although the tuna approached an interrupted light of moderate intensity, they were repelled from the near vicinity at the instant the light flashed either on or off. Future research might be directed profitably at determining the peaction of tuna to reflected light of different quantity and quality originating from moving objects during daylight hours.

Professor Iwao Miyake attempted to discover (1) if tuna produced any sound, and (2) if they could be attracted or repelled by sounds of various frequencies. Using a listening frequency which ranged from about 100 cycles to 70 kilocycles per second, he was able to identify low frequency sounds produced by the sudden movement of the tail of the yellowfin in the tank. This might have some significance in respect to the mechanism of school formation. No sounds produced by the tuna at moderate, high, and supersonic frequencies were detected. In attempting to attract or repel tune by continuous sound stimali, sounds were produced at many frequencies within the 100 cycle to 70 kilocycle rande. No positive results were obtained. However, there were several indications that the tuna might react positively to complex sounds of low frequency. These, and also interrupted sound stimuli over the entire frequency range, might be investigated in the future.

The writer attempted to extend the observations made by Morgan (1951) on the reaction of the abolehole to interrupted direct current in a small wooden tank of seawater. It was found that by progressive shortening of the on-fraction of a cycle at a frequency of 15 cycles per second the downward trend in average current necessary to attract the fish, demonstrated by Morgan, was continued. The relationship between source voltage and electrode size was also clarified. Additional work on the reaction of this species in a small tank of seawater can be undertaken profitably in an attempt to determine the optimum on-fraction and minimum current density for positive attraction. However, this would require a more satisfactory current interrupter than that presently available. After the optimum on-fraction has been determined, the experimental work could be extended to include a study of the reactions of tuna to interrupted direct current in a large volume of seawater such as that of the concrete tank in which the tuna were confined.

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1951. The response of a tropical fish to interrupted direct current and its application to the problems of electrofishing in seawater. M.S. Thesis, University of Hawaii, June 1951: 68 pp.

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PART II

OBSERVATIONS ON THE CHEMORECEPTION OF TUNA2/

by

P. B. van Weel Professor of Zoology University of Hawaii

INTRODUCTION

The experiments to be described in this paper were undertaken during the summer and autumn of 1951 at the Hawaii Marine Laboratory. The author undertook to determine whether tuna have a sense of taste or smell whereby they might be attracted (or repelled) by food substances in suspension or solution. It was hoped to find some component of the food of the fish which acted as an attractant, and which might possibly be prepared in large quantity from some cheap source. If so, it might replace or supplement the live bait which is presently needed for pole and line fishing (June 1951) and which is in short supply in the Central Pacific area.

Originally it was not planned to investigate the reaction of tuna to selected chemicals. However, two such substances were tried at the suggestion of others: asparagine, a possible attractant, and copper acetate, a known shark repellent.

I am very much indebted to Dr. A. L. Tester for his critique and kind help in preparing this paper.

MATERIAL, METHODS AND TECHNIQUE

The experiments were conducted on one or more of two yellowfin (Neothunnus macropterus) and five little tunny (Euthynnus yaito) which were established in a concrete tank (fig. 1) at the Hawaii Marine Laboratory. Normally the tuna were fed a daily ration of tuna flesh from skipjack (Katsuwonus pelamis), yellowfin, or tunny, which they accepted greedily. They also accepted live baitfish (Pranesus insularum), the heads of which had been pinched so that they floundered in the water. They were not observed to feed on a school of baitfish which was present in the tank, probably because they could not develop

^{2/} Contribution No. 23 of the Hawaii Marine Laboratory, University of Hawaii

sufficient speed in the confined quarters to catch them. The tunas accepted dead baitfish, but only when very hungry, and then, with apparent reluctance. They also fed, with apparent reluctance, on marlin (<u>Makaira mazara</u>) flesh after the supply of tuna flesh had been exhausted. The fish in the tank were not fed squid (used in the experiments) although this is one of the food items which they will eat when in their normal habitat (Welsh 1950).

The following substances were prepared and introduced to the tank in a manner which will be described later: I--baitfish water, II-baitfish preparations, III--squid preparations, IV--tuna flesh preparations, V--asparagine and d.l-asparagine solutions, and VI--copper acetate solutions.

The baitfish water consisted of 3 liters of standing seawater in which 50 baitfish had been living for 3 hours. This "conditioned" water was used as a test substance.

The baitfish, squid, and tuna flesh preparations were all made in a similar manner. A quantity (to be reported under each experiment) of the substance was quickly mashed in a blender and the mash was extracted in the refrigerator for 3 hours with twice its weight of distilled water. This "whole" preparation was used in some experiments. In others, the preparation was first fractioned into "clear" and "murky" extracts before being used: the preparation was centrifuged and the supernatant, comparatively clear fluid was diluted to 3 liters with seawater (the clear extract); the remaining debris was suspended in 3 liters of seawater (the murky extract). Variations of or extensions to the above procedure are described under individual experiments. The baitfish were freshly caught; the squid were purchased from the fish market in a frozen condition and thawed just before using; the tuna flesh, from skipjack and yellowfin, was in some cases from freshly caught and in other cases from frozen fish. Marlin flesh, from frozen fish, was also used in a few experiments included under IV.

The asparagine and copper acetate were pure chemicals which were dissolved in seawater in various concentrations as indicated in the individual experiments.

The above substances were introduced to the tank for the most part on the west side at Point A (fig. 2). To eliminate any reactions based on hearing, they were introduced by means of a siphon from a height of about 1 foot, with the rubber tube inlet about 6 inches below the water surface.

Quantitative measurement of the reaction of the fish posed a difficult problem. Normally the tuna would cruise leisurely around the tank in one direction for a long time. At first it was thought



FIG. 2. PLAN OF THE CONCRETE TANK IN WHICH THE TUNA USED FOR EXPERIMENTS IN CHEMORECEPTION WERE KEPT.

that an attractive substance would lure the fish to the rubber inlet and hold them there, but this proved to be the case only with highly attractive substances. With more weakly attractive substances there appeared to be an increase in cruising speed and a tendency to circle closer to the inlet. Accordingly, the reactions were measured in two ways: (1) by determining the time required for the fish to make 10 passes across an imaginary line (AC in fig. 2) drawn across the tank from the inlet, with the fish travelling in either direction (when the fish is circling normally this will be equivalent to the time of 5 complete revolutions), and (2) by counting the number of passes (out of 10) made across the line AB in the near or west half of the tank (when the fish is circling normally there ild be 5 passes across AB and 5 across BC). Therefore, perception of the stimulus would be indicated by a decrease in the time of 10 passes from that of normal, representing an increase in cruising speed and/or a decrease in size of the swimming circle. Attraction would be indicated by an increase beyond 5 in the number of passes across AB, representing one or more circles completely within the near (west) half of the tank. In one case, as indicated later, the substance was siphoned in at C and the number of passes across BC was counted.

It was assumed, and later established, that the reactions would vary with the state of hunger of the fish. To minimize variation from this factor, the tuna were starved for 24 hours before the start of an experiment. However, the state of hunger induced a factor of alertness which could easily result in grave errors: at the approach of an observer the fish became excited, expecting food, and circled close to the observer at increased speed for a considerable period. Tests were not started until the fish became accustomed to the presence of the observer, and a "normal" cruising speed was resumed. As the so-called "normal" cruising speed varied from day to day, it was necessary to establish its value before each experiment. This was done by repeating the timing and counting of the passes until approximately constant values were obtained, with a half-minute interval between successive tests. The substance would then be introduced and the timing test would be continued, still with half-minute intervals between them, until the normal values were again approximated.

As for a long time only one yellowfin was present, many of the results were obtained on this one fish. When more tunas were introduced the schooling instinct became apparent. When two yellowfin and five tunny were present, the two species tended to school separately and to exhibit different reaction patterns. After one yellowfin died, the other joined the tunny, and although a slower swimmer, it attempted to keep up with the school. It showed, therefore, an increased speed of reaction as compared with the results obtained when it was the sole resident of the tank. After four of the five tunny had died, the single tunny schooled with the single, larger yellowfin

(which took the lead) and its reactions were slower than that of the tunny school. These differences in behavior make it difficult to draw general conclusions as to the relative strength of the reaction in the two species.

When the fish schooled, only one member of the school was timed. When both yellowfin and turny were present, the timing tests were conducted alternately on each species.

Finally it should be mentioned that the tuna tended to favor the shady side of the tank, e.g., the east wall during the hours before noon. Before this was realized, many "positive" results were erroneously recorded in morning experiments, with the substance siphoned in at C (fig. 2). To avoid this difficulty, most of the experiments were conducted between 11 a.m. and 12:30 p.m., with the substance siphoned in at A.

RESULTS

The results are shown graphically in figs. 3 to 13. The upper panels show variation in the time in seconds required for 10 passes in either direction across the line AC (fig. 2). The ordinate scale has been reversed in direction so that increase in height of the plotted points indicates increase in cruising speed and/or decrease in the size of the swimming circles, i.e., perception of the stimulus. In the account which follows, the word "cruising speed" has been used to cover the complex behavior pattern measured by the time of 10 passes. The lower panels of figures 3-13 show variation in the number of passes across the line AB; an increase beyond 5 shows that the fish describes one or more complete circles in the near (west) half of the tank, i.e., it indicates attraction to the point of stimulation.

i. Baitfish water

Five experiments were performed with similar results. Only one yellowfin was present. The results of one experiment are recorded in figure 3.

During the preliminary timing, the yellowfin's cruising speed gradually decreased to an approximately constant value. When the baitfish, or "conditioned," water was siphoned in at A, there was no apparent change in either cruising speed or in the number of passes across AB. Similarly, when the baitfish water was siphoned in at C, there was no apparent change in cruising speed or in the number of passes across CD. The experiment does not demonstrate the false "positive" attraction which may result from the tuna's preference for the shady side of the tank, although it was shown in other preliminary





experiments. It may be concluded from this and the four other experiments that baitfish sater has no apparent attractive or repellent effect.

Figure 3 also shows the results of a control experiment in which seawater was siphoned in at A. Again, there was no noticeable or measurable reaction of significance.

ii. Baitfish preparations

In several experiments, one of two schooling yellowfin was timed in preliminary tests, on introduction of the clear extract of the baitfish preparation, and on introduction of the murky extract of the baitfish preparation. The results of only one experiment are recorded in figure 4_9 in which 151 grams of baitfish (wet weight) was used.

On introduction of the clear extract, there appeared to be an increase in cruising speed and an increase in the number of passes, particularly in Tests 10 and 11. The mean decrease in time for 10 passes was from 112.3 seconds for the preliminary tests to 96.3 seconds for the tests with the clear extract. The difference is not statistically significant. 3/

On introduction of the murky extract there was at first a slight increase and then a considerable decrease in cruising speed. The initial increase might be construed as a reaction to the substance, but not necessarily as an attraction, as the number of passes across AB did not increase.

The other experiments yielded even less evidence of attraction to the baitfish preparations. It may be concluded that this substance has either very slight attractive properties or none at all.

A simple test of the significance of the difference between the mean times is not necessarily informative in experiments of this nature as (a) during the preliminary timing there may be a gradual decrease in cruising speed as the fish become accustomed to the presence of the observer, and (b) during siphoning, there may be at first no change, then an increase, and finally a decrease in cruising speed (and number of passes) during the times in which the substance is entering the tank, spreading over a small volume of sea water near the inlet, and gradually dispersing in smaller concentration farther and farther from the inlet. Unfortunately the data are not sufficiently extensive for more detailed statistical analysis even if a procedure could be designed to handle this complex situation.





iii. Squid preparations

Experiments with squid preparations, either whole or divided into clear and murky portions, yielded no evidence of the presence of attractive substances even when comparatively concentrated preparations were used.

As the squid is a normal food of the tunas and as it will expel ink when pursued, the possibility exists that the combined stimuli of sight and smell might evoke a response whereas either one, acting alone, would not. To test this possibility, Preparation A was made in the following manner. Fresh (thawed) squid (204 grams) was extracted without centrifuging, and to this was added the ink of a freshly speared octopus (Polypus marmoratus). The material, when siphoned into the tank, formed a darkish cloud which was visible from above. Two schooling yellowfin were present, one of which was timed. The results are shown in figure 5.

In preliminary Tests 1 to 7, the tuna were attracted by the presence of the observer but settled down to normal activity during Tests 8 to 13. While introducing the preparation, there was an increase in cruising speed during Tests 16 and 17. This, however, cannot be regarded as a reaction to the substance as the behavior was not repeated in a second experiment. Moreover, the number of passes across AB remained constant at 5 throughout Tests 14 to 21, indicating no attraction.

Although the cloud formed by Preparation A was plainly visible, it was not particularly dark when the preparation became diluted in the tank. As the walls of the tank were also dark, there remained the possibility that the contrast was not sufficiently sharp. Accordingly, Preparation B was made in a similar manner (365 grams of squid) except that India ink was added in such quantity as to produce a "pitch" black cloud in the tank. Two yellowfin and one tunny were present, all schooling together. The results are included in figure 5.

As indicated in the preliminary tests, for some unknown reason the fish were restless and cruised around at a relatively high speed. When Preparation B was siphoned into the tank, the tuna avoided the black cloud, remaining close to the south wall (fig. 2), and not crossing the line AC. The cloud diffused across the tank and gradually drifted towards the south wall. As it approached, the yellowfin became more and more excited, and finally darted through it, thereafter remaining in the up-stream portion of the tank until practically all of the cloud had dispersed through the overflow. The tunny showed an entirely different behavior, appearing to be undisturbed by the presence of the black cloud. It cruised into and cut of the cloud, maintaining an approximately constant speed.





It may be concluded that the squid preparation either of itself or in combination with squid ink or India ink had no attractive effect on the time. The ink, when concentrated, appeared to have a repellent effect on the yellowfin, but this was probably a visual reaction. It is unlikely that the repellent effect could have been produced by some chemical in the India ink as the carbon particles are usually stabilized in suspension by gelatin, and gelatin has no effect on the behavior of the fish as shown by a control experiment with a 1.0 percent solution.

iv. June flesh preparations

During most of the experiments with tuna flesh preparations, two yellowfin and five tunny were present in the tank, forming two distinct schools with markedly different cruising speeds. Only one individual (not necessarily the same fish throughout the experiment) of each school was timed. The timing tests on each species were conducted alternately.

A. In the experiment portrayed in figure 6 the whole extract of skipjack flesh (200 grams) was used, without centrifuging. The tunny, in particular, were attracted by the observer's presence and did not slow down and extend their course over the whole tank for a considerable period of time. They appeared to have an exciting effect on the yellowfin, the cruising speed of which increased during the preliminary trials. While the preparation was being siphoned into the tank, there was no reaction during the first few seconds until the tunny neared the inlet. Then, however, the tunny started milling around the inlet excitedly. The yellowfin, also, soon showed this same reaction. One of the latter actually snapped at the rubber inlet. All fish took shreds and small pieces of the extracted flesh into their mouths, but invariably spat them out agoin. Apparently this material, after extraction, is tasteless and impalatable to the fish, even though they normelly were fed tuna flash while in captivity. However, other fish in the tank, (a puffer, Tetrodon hispidus (?) and a few manini, (Acanthurus sandvicensis) did eat the extracted material. Although it required only 3 to 5 minutes to siphon the 3 liter preparation, the general excitement of the tuna lasted for some 10 to 15 minutes. The timing (Tests 17 to 25 for yellowfin and 18 to 26 for tunny) was then resumed as the fish grew calmer and started cruising again.

B. A series of experiments was next conducted in which the tuna flesh preparation was fractioned into clear and murky portions. As these all gave similar results, only one is recorded in detail in figure 7, in which 130 grams of skipjack flesh was used.

During the preliminary tests, the speed of the tunny school gradually decreased, whereas that of the yellowfin remained fairly constant. When the clear extract was introduced, there was a difference in behavior of the two species. The tunny started milling around the









inlet after about 30 seconds. They became greatly excited and often bit into the rubber tube. Some 10 minutes after siphoning started, the excitement subsided and some 5 minutes later they resumed their cruising. The yellowfin were markedly excited and were definitely attracted to the inlet, which they snapped at occasionally. However, they did not mill around the inlet, but rather increased their cruising speed once they sensed the clear extract. It should be emphasized that this clear extract was invisible in the tank.

After 45 minutes, during which period the fish became calm and resumed their normal cruising speed, the murky portion was introduced. In the experiment recorded in figure 7, the tunny's speed was high during the first minute of siphoning but decreased thereafter. From this and other experiments it was concluded that the murky portion had no apparent effect on the cruising speed of either the yellowfin or the tunny. Both species, however, were attracted by the whitish color of the murky extract and by the shreds of flesh contained in it. They snapped at the shreds but did not swallow them. This attraction is indicated by the increased number of passes across AB in Tests 36 to 40.

C. One experiment using clear and murky extracts of skipjack flesh (120 grams) was performed after the fish had been recently fed. Neither the cruising speed nor the number of passes gave any indication of a positive attraction. This was expected from observations of their feeding activity in the tank. On throwing food to them, at first they take it greedily, milling around at increased speed, soon they react more slowly as their hunger is satisfied, finally they ignore the food.

D. Since it was established that the clear extract contains the attractive factor(s), an attempt was made to determine whether this was contained in the "fat" (petrol ether soluble) or "protein" (residual) parts. The fat fraction was obtained by shaking the clear extract with petrol ether and separating the latter from the residue or protein fraction. After evaporation of the petrol ether in the refrigerator (where the protein fraction was suspended in the same amount of distilled water as that of the protein fraction. Both fractions were diluted with seawater to 3 liters. In the experiments which are discussed below, one yellowfin and five tunny were present. All fish tended to school together, with the yellowfin trailing.

In one experiment, the results of which are shown in figure 8, yellowfin flesh (200 grams) was used, yielding 350 cubic centimeters of the clear portion. This was shaken with 75 cubic centimeters of petrol ether. During the preliminary trials, the fish appeared to be hungry, and were excited by the presence of the observer. The protein fraction was siphoned in first. During the first 2 minutes there was





no reaction, but then the fish swam rapidly in small circles around the inlet. The reaction, particularly of the tunny, was clearly positive, although not as violent as with the normal clear extract. The yellowfin behaved similarly to the tunny, but might have been influenced by their behavior. None of the fish snapped at the inlet. After normal cruising of the tank was resumed, the fat fraction was introduced. There was no detectable reaction.

A second experiment illustrates the behavior somewhat better. as the reaction was somewhat less pronounced and the timing tests could be continued throughout. In this, yellowfin flesh (180 grams) was again used, yielding 225 cubic centimeters of the clear solution which was shaken with 50 cubic centimeters of petrol ether. Both species schooled together, with the yellowfin trailing. The results are shown in figures 9 and 10. As soon as the protein fraction was introduced, the fish were attracted and cruised in small circles at the inlet. The yellowfin "sniffed" at the tube during Tests 15 and 19. and the tunny both "sniffed" and snapped at the inlet during Test 18. After waiting 25 minutes, the fat fraction was siphoned in. Unfortunately when siphoning was started, two onlookers appeared whose presence attracted and excited the fish (Tests 33 to 40). When they went away, the normal cruising speed of the fish was resumed. Except for the disturbing presence of the onlookers, the results are similar to those in the previous experiment. It appears that the attractive substance is located in the protein rather than the fat fraction of the clear extract of tuna flesh.

E. A series of experiments was next performed to determine the extent to which the tuna flesh preparation could be diluted and still retain its quality of attraction. In these the whole preparation was used, but it was made up in various dilutions. It should be kept in mind that the concentrations given are those siphoned into the tank, and not those in the tank. The concentration in the tank would be difficult to determine as it does not remain constant because of diffusion.

In the first experiment, one yellowfin and five tunny were present schooling together. The whole preparation of yellowfin flesh (156 grams, yielding 300 cubic centimeters of whole extract) was divided into the following quantities and each was diluted to 3 liters with seawater: (1) 10, (2) 50, (3) 90, and (4) 150 cubic centimeters. The results are shown in figures 11 and 12. When (1) was siphoned in, there was an increase in cruising speed but no definite attraction to the inlet. When (2) was siphoned in, there appeared to be a definite attraction as both the yellowfin and tunny swam in small circles near the inlet during the siphoning process. When (3) was siphoned in, the reaction was similar, but for some unknown reason it was less pronounced than in (2). When (4) was siphoned in, the reaction soon became very pronounced, both species milling around near the inlet at high speed for about 5 minutes.



FIG. 9. GRAPHS ILLUSTRATING THE REACTION OF ONE YELLOWFIN TO "PROTEIN" AND "FAT" FRACTIONS OF CLEAR EXTRACT OF YELLOWFIN FLESH PREPARATION.

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FIG. 10. GRAPHS ILLUSTRATING THE REACTION OF FIVE TUNNY TO "PROTEIN" AND "FAT" FRACTIONS OF CLEAR EXTRACT OF YELLOWFIN FLESH PREPARATION.









It was observed in this, as in many other experiments, that before the substance was introduced, the two species formed one compact school, with the yellowfin trailing; but when an attractive substance was introduced, and the fish became excited, each species reacted at different cruising speeds, thus breaking up the compact school formation.

In a second experiment, one yellowfin and two tunny were present, the yellowfin leading the school. The whole preparation of yellowfin flesh (150 grams, yielding 300 cubic centimeters of whole extract) was divided into the following quantities, each of which was diluted to 3 liters with seawater: (1) 5, (2) 10, (3) 50, and (4) 235 cubic centimeters. Three of the tunny had died and the remaining two seemed listless when this experiment was performed. Consequently, strong reactions were not expected. The results were similar to those of the previous experiment although the reactions were not as pronounced. There was an indication that the tunny sensed the most dilute (1) solution in that they often hesitated at the inlet but then quickly increased their speed so that they caught up with the yellowfin. Both the tunny and the yellowfin made more passes across AB than BC, indicating attraction。 With (2), the stimulus was apparently strong enough to cause the tunny to leave the yellowfin and react independently. Again. an increase in cruising time and number of passes for both species indicated attraction. With (3), the tunny frequently remained near the inlet, and only occasionally joined the cruising yellowfin. Again attraction was indicated in both species. With (4) both yellowfin and tunny showed a pronounced increase in cruising speed and in number of passes across AC. However the fish did not mill around the inlet as in the previous experiment, even when the remainder of the whole preparation was poured into the tank.

F. The objection might be raised that attraction to the tuna flesh preparation occurs because the tuna were conditioned to this kind of food, and that a similar reaction would not necessarily be obtained with "wild" fish. This possibility cannot be denied, although it is shown in the experiment to follow that the reaction was also obtained with marlin flesh. The only survivor, the yellowfin, had not yet been fed this material so it could not have been conditioned to it.

Unfortunately, the yellowfin was in goor condition and did not show pronounced reactions to its food. Accordingly it was considered that even a weak reaction would be evidence of an attractive substance. Several experiments were performed with similar results. Only one is recorded in figure 13 in which 260 grams of marlin flesh was used as a whole preparation.



MARLIN FLESH PREPARATION.

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During preliminary Tests 1 to 10 the cruising speed was erratic and the fish often approached the observer, apparently expecting food. The cruising speed became more regular during Tests 11 to 16. The marlin flesh preparation was then siphoned into the tank. There was an initial increase in cruising speed, followed by a decrease, during which time (Trials 18 and 19) the yellowfin hesitated near the inlet apparently sensing the substance. Following this, the cruising speed increased again, and then gradually decreased. The number of passes across AB also increased during the siphoning period, indicating attraction. From these, and similar results in other experiments, it is concluded that the yellowfin, even though in poor condition, was attracted by the marlin flesh preparation.

v. Asparagine and d.l-Asparagine

One yellowfin and five tunny were present in the tank during several experiments with these chemicals. Solutions ranging from 0.1 to 1.0 percent were used. As no reactions whatever were observed, the results are not included here. It may be concluded that these substances were not noticed by the tuna.

vi. Copper acetate

The question arose as to whether copper acetate might be used to repel sharks without repelling tuna during long-line fishing operations. It was suggested that some information on this point might be obtained by determining the reaction of tuna in captivity to this chemical. While it was not possible to compare the reactions of the tuna to that of sharks, it was possible to compare them with the reactions of a few manini and baitfish which were present in the tank.

The following concentrations of copper acetate were used: 0.1, 0.2, 0.5, and 1.0 percent. For each experiment, 3 liters of these solutions were siphoned into the tank at A, at C, or at both A and C together, with the solutions flowing down-stream. A part of the tank was thus kept clear of the solution. Stronger concentrations were not used for fear of injuring or killing the tuna. When siphoned into the tank, the solutions were clearly visible from above as a bluish cloud. The actual concentration of copper acetate in this cloud was not determined; it changed rapidly as the cloud diffused.

The behavior of the tuna was similar at all concentrations of the solution and the reactions differed only in degree. The first fish to exhibit reactions were the manini. They swam up-stream and remained near the seawater inlet for the duration of each experiment. The baitfish also avoided the down-stream part of the tank into which the solution was being siphoned. In general, the yellowfin and tunny cruised around the cloud of copper acetate solution but did not enter it. As the cloud diffused, both species kept to the up-stream part of the tank. After the cloud had diffused until it was barely visible, the tuna eventually entered it. With the stronger solutions it took a proportionately longer period of time for the tuna to return to the down-stream part of the tank. Apparently there is a critical concentration above which there is a repellent effect. This, however, is not too marked as in one experiment (the only one with this reaction) both the yellowfin and the tunny swam right through the cloud (0.1 percent, or weakest solution) from the start of siphoning. Despite this instance, it may be concluded that copper acetate solution has a repellent effect on tuna, although its action is not as pronounced as in the case of manini and baitfish.

DISCUSSION AND CONCLUSIONS

As the tuna are predacious fish, it might be expected that the most important sense involved in feeding would be the eyesight, followed perhaps by hearing. However other senses, as for example the chemical sense in its widest meaning, cannot be excluded on an <u>a</u> priori basis. The experiments reported in this paper show that a chemical sense is present, and this indicates that it may play some part in feeding. It is impossible to say whether the chemical sense is smell or taste or, in other words, whether sense organs in the mouth or the nose are being stimulated. However, both senses are usually well-developed in fish as other investigators of freshwater and marine fishes have shown (Adrian and Ludwig 1938; Berghe 1929; Copeland 1912; von Frisch 1941; Greene 1925; Göz 1941; Hasler and Wisby 1949; Huttel 1941; Klenk 1930; Neurath 1949; Parker 1910, 1911, 1913, 1922; Scharrer, Smith, and Palay 1947; Sheldon 1911; Strick 1924, 1925; Trudel 1929; Walker and Hasler 1949; Wrede 1932).

The experiments of Göz (1941) and Wrede (1932) showed that the skin of fish secretes a substance which can be perceived chemically not only by fish of the same species but also by fish of other species, and that the substance could be recognized by smell. It was thought that the same might be true for tuna, i.e., that they could smell the presence of other fishes. However, this was not the case, as shown by the negative experiments with "conditioned" water in which baitfish had been living.

Von Frisch (1941) found that the skin of injured minnows (Phoxinus laevis) gives off odoriferous substances, probably purinor pterin-like which cause alarm reactions in the same and related species. It was thought that an alarm or repellent stimulus to the prey might be an attractant stimulus to the predator, i.e., that baitfish or squid preparations might attract the tuna. The experiments reported here brought quite unexpected results to light: the tuna reacted not at all, or only weakly, to extracts of squid (a normal food) and of baitfish (a food which they will take when thrown from a fishing boat), whereas they did react quite strongly to extracts of tuna flesh. This attraction was based on chemical stimulation rather than on sight, as proven by experiments with the clear, centrifuged portion of the tuna flesh preparation in which the invisible fluid attracted the fish. The tuna were attracted to the debris by sight, but they did not accept the particles as food, even when taken into the mouth. It was shown that an extract equivalent to 5 grams of tuna ilesh in 3 liters of seawater, siphoned into the tank, was sensed by the tunny, and that in one experiment, at least, an extract equivalent to 25 grams of tuna flesh in 3 liters of seawater was positively attractive to both yellowfin and tunny. Extracts of marlin flesh also gave positive reactions, showing that the response in the tuna flesh experiments was not conditioned by the food.

Experiments with "fat" and "protein" fractions of the aqueous extract of tuna flesh showed that the attractive substance was in the latter. This is in contradiction to the results of Allison and Cole (1934) who found that fatty acids had an effect on both freshwater and marine fishes. It must be left for future research to determine what part of the protein-containing fraction is the actual attractant. Von Frisch's (1941) experiments might be recalled in this connection, suggesting that such substances might be purin- or pterinlike.

The tuna displayed no positive reaction to either asparagine or d.l-asparagine solutions.

Experiments with copper acetate, a well-known shark repellent (Whitley and Payne 1947) showed that this substance has a repellent action on tunas. However, they were not as sensitive to this chemical as other fish (manini and baitfish) which were also present in the tank. Only a few experiments with relatively weak solutions were conducted because of danger of harming the tuna.

It should be emphasized that the experiments were conducted with tuna in captivity, rather than in their normal habitat, and that the reactions in the latter might be different. It should also be emphasized that when both yellowfin and tunny are present in the tank, there is an interaction which affects both cruising speed and schooling pattern. For this reason, caution must be exercised in comparing the intensity of the reactions of the two species. In general, however, the reactions of the tunny seemed to be considerably more pronounced than those of the yellowfin, indicating a greater sensitivity to the attractive substances.

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PART III

OBSERVATIONS ON THE REACTION OF TUNA TO ARTIFICIAL LIGHT

by

Sidney C. Hsiao Associate Professor of Zoology University of Hawaii

INTRODUCTION

The response of fish to visual stimuli was studied as early as 1880 by Kühne and Sewall. Since that early date, a great deal of information has been accumulated on the structure and function of the piscian visual organ, and on the question of color vision in fish. a subject of discussion and dispute amoung physiologists. The literature on color vision in fishes has been critically reviewed by Warner (1931). Wall's (1942) monograph dealing with the adaptive radiation of the vertebrate eye brings the literature review up to 1941. No further review has appeared over the past 10 years. Nearly all the experiments on piscian vision, both achromatic and chromatic, were done on favorable laboratory specimens. which were hardy and of suitable size for indoor tanks. As far as can be ascertained, no experimental work on the physiology of vision in tuna has been reported; none is included in a recently published bibliography on the biology of the Pacific species (Shimada 1951). Field observations made by amateurs and professional fishermen have been accumulated for some time, but conclusions based on them are badly in need of verification.

It is the purpose of these studies to discover the pattern of reaction of tuna, established in captivity, to different quantities and qualities of light stimuli. The former is concerned with the intensity and duration of light stimulation, and the latter with the frequencies of the light used, that is, the different portions of the visible spectrum which are involved. The success of Tester (1952) in capturing several species of Pacific tuna, transporting them to shore, and keeping them alive in the confined space of an outdoor pond and tank, makes it possible to experiment with these oceanic species under controlled conditions. It was hoped that attraction or repulsion stimuli would be discovered which might have some use in explaining the behavior of tuna in their natural habitat, and perhaps in suggesting new or improved methods of capture.

The work was undertaken during the summer of 1951 at the Hawaii Marine Laboratory under the general direction of Dr. A. L. Tester, University of Hawaii, whose assistance is gratefully acknowledged.

^{4/} Contribution No. 24 of the Hawaii Marine Laboratory, University of Hawaii, Honolulu, T. H.

The fish used in these studies were one or two yellowfin (<u>Neothunnus macropterus</u>) and five little tunny (<u>Euthynnus yaito</u>), which were confined in a concrete tank (**T**ester 1952).

A general plan of the tank is shown in figure 14. In this A_{g} B, C, and D are four metal baffles, painted white; the longer baffle D houses optical equipment E. The seawater inlet is indicated by G and its exit by H, while L and M show the positions of two 60-watt electric light bulbs which were lit each night from dusk to dawn. The dotted line NO indicates the position of a cord placed across the tank about 1 foot above the water surface to mark off the northern one-quarter of the tank. It was fastened to the slat railing which surrounded the tank. The feeding station is shown by F.

The instrument box E, shown in detail in figure 15, is designed to carry a source of light, a set of light filters, and shutters with variable speed, submerged so as to send a beam of light of desired frequency horizontally under the surface of the water from one end of the tank to the other, and to prevent seawater from coming into contact with the instruments. The box is 24 inches long, 18 inches wide, and 24 inches deep, made of galvanized iron sheets soldered at the joints, and held rigid by a wooden frame. At first a carbon arc lamp with a series of optical lenses was used as the source of light. However, the inconvenience of having to change the carbon pencil every half-hour or so led to its replacement by a projection lantern. The lantern (L in the diagram) consists of a 500-watt Mazda incandescent lamp placed in front of a concave mirror and behind a series of optical lenses which concentrate the light into a narrow beam of about $l\frac{1}{4}$ inches in diameter at the glass window W. The lantern is connected to the 110-volt, 60 cycle power line through a powerstat (variable transformer) which has the following specifications: output range, 0-135 volts; maximum amperes, 7.5; output KVA, 1; frequency, 50/60 cycle. A metal frame 1 inch larger all around than the 3×4 inch glass plate used as a window W is soldered on the outside of the box over the 3-inch square opening to hold the glass in place, while a patented caulking compound forced into the space between glass and frame successfully makes the window water tight. On its way to the window the light beam passes through a shutter S which is a circular disc of aluminum carrying four openings, each l_{4}^{1} inches in diameter and placed equidistally one in each quadrant. The axis of the disc is mounted against a friction disc F connected with a 3-inch pulley P. By changing the point of contact between the friction wheel and a fixed ring on the axis of the shutter, the speed of rotation of the shutter can be changed at will from no motion, when the point of contact is at the center of the friction wheel, to maximum speed, when the contact is at its rim. By selective use of either one, or two, or all four openings in the shutter, the rate of interruption of light can be increased three-fold. A



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FIG. 15. PLAN OF INSTRUMENT BOX HOUSING LIGHT-PRODUCING AND OPTICAL EQUIPMENT.

secondary shutter E, placed across the lens system of the projection lamp, can be manipulated by hand to produce variable and irregular flickering. The pulley P is driven by a 1750 RPM, $\frac{1}{4}$ -HP motor through a series of reducing pulleys and V-belts not shown in the diagram. In front of the shutter S, behind window W, a Masonite light block M and 3 Kodak light filters A, B, and C are mounted in such a way that each can be easily pushed into the path of the light beam.

The three Kodak filters are listed as "Series VII Wratten filter A, #25; B, #58; and C5, #47," respectively. Their transmission characteristics were examined with a Beckman Model B spectrophotometer with the results shown in figure 16. The #25 red filter transmits a band of light with wave lengths between about 590 and 700 m μ , the #47 blue filter between about 360 and and 530 m μ , with maximum transmission at 440 m μ , and the #58 green filter between about 470 and 615 m μ , with maximum transmission at 525 m μ .

The intensity of illumination was controlled by use of the powerstat when using the lantern, or by changing electric bulbs from one wattage to another by methods to be described below. As lights of different frequencies have different intensity and different penetrating power through water the intensity was measured in situ by means of a photoelectric cell and an attached microammeter. The microammeter was calibrated under standard conditions $\frac{6}{2}$ in terms of foot-candles and the calibration curve was used in determining the intensities of the different lights used as stimuli.

A piece of heavy canvas was draped over the instrument box E to protect the apparatus against the weather and to serve as a blind for the experimenter. An opening about 2 inches square was cut in the canvas as an observation "peek hole."

To contrast the effect of a single beam of light with that of night lights used in certain commercial fisheries, incandescent electric light bulbs were adapted for submergence under water and for carrying a frame for glass filters of different colors. To produce different quantities of light for stimulation, 40-, 60-, 100-, and 200-watt bulbs were used, while to produce intermittent light a practically noiseless mercury switch was introduced into the line. Commercial colored electric bulbs (similar to those

^{5/} The Kodak catalog lists this filter as transmitting two portions from the spectral band: a major portion between 480 and 640 mu and a minor one between 670 and 700 mu, with maximum transmission at 520 mm

It is a pleasure to thank Mr. R. Oberdorfer of the Physics Department for his help in the calibration.



FIG. 16 TRANSMISSION CHARACTERISTICS OF KODAK WRATTEN FILTERS

used for decoration) were also employed. In all experiments, the bulbs were hung on the tank side of baffle D (fig. 14) and were practically or completely submerged.

Preliminary studies showed that during daylight hours the artificial light; in contrast to sunlight, was too weak to elicit definite response from the fish. Furthermore, even at late afternoon, the reactions were irregular because of various disturbances about the tank. Therefore, all the experiments were performed after dusk or at night and at times when the tide was sufficiently low to insure clear water in the tank for comparable light penetration.

As the tuna fed regularly from the western side of the middle of the southern half of the tank (at F in fig. 14), all light stimuli were applied from the northern edge of the tank and only when a tuna entered into a specifically selected area or field in the northern one-quarter of the tank would its reaction be recorded. This field is defined by the following method: by applying one's eye to the observation opening in the canvas described above, the observer can bring the top edge of this 2-inch square hole to coincide with the twine NO (fig. 14), and the middle point of its right side with the shadow of the electric light L. With a fixed line and fixed point so selected, a square shown by the area PGRS in figure 14 is determined. This area was used as the field of observation. Since this area is immediately in front of the source of light and away from the feeding site, and traversed by the light beam, entrance by tuna into this area as they travel toward the light was taken as a criterion of reaction. Rec rds were made of the number of times during an experimental period that the tuna swam into this field, and also of the pattern of movement. An experimental period, the time during which a particular stimulus was applied, was arbitrarily fixed at 5 minutes.

Before starting a series of experiments, preliminary observations were made of the behavior of the fish. When more than one was present they tended to form a school and cruised slowly about the tank, occasionally passing between the observation area and the baffle D (fig. l4), but more often circling in the southern three-quarters of the tank. Only occasionally during a period of about 5 minutes would they enter the observation area: the number of entrances was usually O_9 occasionally 1_9 and more rarely 2 or 3. This was taken as control condition, and care was taken that this condition was present before the start of each experiment, although the number of entrances was not always recorded nor included in the tables which follow. At times the fish were unduly disturbed by extraneous factors such as the presence of visitors or an urge to chase a school of baitfish which was present in the tank. Experiments which were attempted under these conditions gave erratic results and were rejected.

All experiments were performed while the tank was illuminated by two 60-watt bulbs (L and M in fig. 14) which were suspended about 6 feet above the water's surface. These lights were not extinguished during the experiments for fear of frightening the fish and causing them to ram the walls. They served to illuminate the tank sufficiently for observations to be made of the behavior of the fish in waiting for the establishment of control conditions and during the experiments.

RESULTS

Several series of experiments were conducted between August 20 and September 18, 1951, each on a different night and therefore each under slightly different environmental conditions. They are discussed below in the order in which they were performed.

Series i--Reactions to white ?/ and colored beams of light from a carbon arc lamp.

The results of experiments with a continuous horizontal beam from a carbon arc lamp are shown in table 2. The estimated intensity of the beam, using white light, was about 450 foot candles (the apparatus for measuring the intensity was not available at the time of the experiments). Only one yellowfin was present in the tank.

After control conditions were established, the Masonite shutter was slid from the window as quietly as possible and the beam was allowed to penetrate the tank for 5 minutes. During this period, the tuna entered the field 14 times in Experiment 1 and 18 times in Experiment 2. The pattern of movement was fairly regular. The fish swam across the beam of light at the south end of the tank, then turned around, swimming parallel to but outside of the beam towards the source. After entering the field at the north end of the tank, it again turned around, passing through the light beam with its one side and eye illuminated. This is considered to be a definite tropistic reaction. At times, attraction was also indicated when the fish, on leaving the field, described a small circle in the northern half of the tank, and re-entered the field. When the light was turned off, the swimming pattern was maintained for but a minute

The word "white" is used in the sense that no light filters were employed to regulate the emitting light beam's frequency.

Remarks	Regular pattern of movement	E	Pattern similar to above	E	£	Entrances in first 2 minutes	Regular pattern of movement
Number of entrances to field in 5 min.	14	18	13	14	17	m	13
Color	white	white	red	red	red	0 9	green
Light	Carbon arc	Carbon arc	Carbon arc with filter	Carbon arc with filter	Carbon arc with filter	8	Carbon arc with filter
Experi- ment_no.	Ч	2	ŝ	4	Ś	Control	9

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Table 2.==Series 1==Reactions of tuna to white and colored beam of continuous light from carbon arc lamp.

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or so. Thus, in the control condition, listed in table 2, the fish entered the field 3 times during the first 2 minutes, but not thereafter.

In Experiments 3 to 6, colored filters were used. With the red filter, the tuna entered the field 13_{\circ} 14, and 17 times in three separate experiments. With the green filter, the tuna entered the field 13 times in one experiment. Under control conditions, as before, there were only 3 entrances. The pattern of movement was similar to that with white light.

Apparently both white and colored continuous light from the horizontally projected arc-light beam caused a similar tropistic movement. Unfortunately, experiments with a blue filter were interrupted by noise emanating from nearby buildings.

Series ii--Reactions to radiating light from bulbs

The results of experiments with continuous and interrupted radiating light from a plain 200-watt (215 foot candles) and a painted 200-watt (106 foot candles) electric light bulb are shown in table 3. The painted bulb, with a slight orange hue, was used because it happened to have been equipped with a waterproof socket; it had been used previously in night lighting for fish larvae. Two yellowfin and five tunny were present.

Experiments 1 to 5 were performed with continuous light from the two bulbs. In Experiments 1 and 2, which were made at dusk (6:40 and 6:55 p.m.) there was no positive reaction, the tuna entering the field fewer times during the experiment than during control conditions. The negative results may have been due to the fact that it was not quite dark when the experiments were made. A half-hour later, positive tropism was shown in Experiments 3 and 4 with 14 and 12 entrances into the field as compared with 4 under control conditions. At times the yellowfin entered the field by themselves; at other times they joined the school of tunny, which entered as a unit. In Experiment 5, with the plain bulb, there were only 6 entrances, but this may have been due to interference caused by the approach of onlookers to the south wall of the tank towards the end of the 5-minute test period. It is concluded that, after dark, the tuna show tropistic response to radiating light from bulbs with intensities of 215 and 106 foot candles.

In Experiments 6 to 10, the plain and painted bulbs were interrupted at a rate between 60 and 75 times per minute, with the light and dark period of the same duration. The number of entrances with the painted bulb (11, 11, and 7) was greater than with the plain bulb (0 and 4). The latter did not differ appreciably from control conditions. However, observations

Table 3Series ifReactions of tuna to continuous and interrupted radiated white light	Remarks	One entrance by yellowfin	One entrance by yellowfin, one by tunny school	More entrances than normal	Entrances in schools	4 entrances by yellowfin _s 8 by mixed school		Onlookers approached tank from south	Entrances mostly in schools	Turned away at border with change of light	Entrances mostly in schools	£	Entrance only during first 2 minutes
and inter	Number of entrances to field in 5 min.	Ч	ъ	¢	14	12	4	9	11	0	11	7	4
con tinuous	Intensity (foot- candles)	215	106	0 0	215	106	0	215	106	215	106	106	215
ons of tuna to	Rate of interruption per minute	0 0	8 0	0 C	8	i O	0 0	9 8	60	02	70	75	75
le 3Series ilReacti	Light	Plain 200-watt bulb	Painted 200-watt bulb	0 0	Plain 200-watt bulb	Painted 200-watt bulb	0 8	Plain 200⇔watt bulb	Painted 200-watt bulb	Plain 200-watt bulb	Painted 200-watt bulb	Painted 200⇔watt bulb	Plain 200-watt bulb
Tat	Experi- ment no.	Ч	2	Control	σ	4	Control	Ś	9	5	to	6	10

24.0 Table 3.--Series it --Reactions of tune to continuous

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of the behavior of the fish showed that with the plain bulb the fish were attracted to the source, but with each abrupt change between light and darkness, they were frightened away if near the source, e.g., near the south edge of the field. On turning away from the light source, they swam faster than usual and broke the school formation. Thus, the tuna appeared to be attracted to lights of 106 and 215-candle power when interrupted about once per second, but were repelled when close to the stronger source, by sudden interruption or application of stimuli.

Series iii-Reactions to radiating light from bulbs

In these experiments, summarized in table 4, an attempt was made to repeat the results of Series ii, using the painted 200-watt bulb and a 100-watt bulb of the same luminous intensity (106 foot candles), and a 60-watt bulb with a lower intensity (70 foot candles). One yellowfin and five tunny were present.

Experiments 1 to 4 demonstrate a positive tropism to continuous light at both intensities, thus confirming similar results in Series ii. In every case, the number of entrances under experimental conditions was considerably greater than under control conditions.

In Experiments 5 to 9, the light was interrupted regularly at various rates, and also irregularly. In the latter, the time of interruption was varied during the course of the experiment to observe the effect of the change from light to dark (and vice versa) on the pattern of movement. With the 100-watt bulb there was positive tropism at regular rates of 70 and 100-120 times per minute with the on and off periods equal, and also with irregular rates, with no appreciable difference in the strength of the response between the regular and irregular rates. Similarly there was positive tropism at a regular rate of 6 times per minute, with the on period 0.5 and the off period 9.5 seconds. These results confirm and extend the observations included in Series ii. With the 60watt bulb, however, there was no definite attraction to irregular interrupted light, the tuna entering the field only twice. This lack of response is possibly associated with the low intensity of the bulb (70 foot candles).

Series iv--Reactions to radiating light from white and colored bulbs of low intensity.

In these experiments sources of light of smaller luminous intensity than that of the overhead lights were used with continuous and intermittent stimulation. One yellowfin and five tunny were present. The results are given in table 5.

Table 4.=-Series iii.=-Reactions of tuna to continuous and interrupted radiated white light	Rate ofNumber ofRate ofIntensityinterruption(foot=to fieldto fieldper minutecandles)in 5 min.Remarks	t bulb 106 14 4 entrances at border, 10 in field	er l One entrance by yellowfin at border	att bulb 106 13 Entrance in schools; definite tropistic reaction	bulb == 70 13 m	ee 5 3 entrances during first minute	att bulb 106 13 Entrance in schools, except twice by yellowfin	9 0 0 0 0	70 106 IO	0 0 0 0 0 0 0 0	100=120 106 13 E	irreg。 70 2.		irreg. 106 12 I	t bulb 6* 106 13 field "
able 4Series iiiReactions of	Light	Plain 100-watt bulb	8 0 0 9	Painted 200-watt bulb	Plain 60-watt bulb	0 0 9	Painted 200-watt bulb	0 0 0	Plain 100-watt bulb 70	0 9 0 8	Plain 100-watt bulb 100-120	Plain 60∝watt bulb irreg.	0 0 0	Plain 100-watt bulb irreg.	Plain 100-watt bulb 6*
Тя	Experi- ment no.	r-1	Control	5	e	Control	-7 4	œ Control	Ś	Control	6	٢	Control	80	6

In all other experiments on and off periods were equal. *0 5 seconds on 9.5 seconds off.

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D	r of nces eld <u>min</u> . <u>Remarks</u>	Entrances by yellowfin in second and fourth minute				Entrances once by solitary fish, twice by schools							
	Number of entrances to field in 5 min.	2	0	0	0	ς	0	0	0	0	0	0	0
	Intensity (foot- candles)	47	0 0	47	8 8	215	47	47	0 0	8	3	6	75
radiated white light	Rate of interruption per minute	0 0	89	89	8 0	0 0	60	Я	0 P	9.0	0 0	0 9	0 9
radiated	Color	white	0 0	white	0 0	white	whi te	white	6 0	red	yellow	green	amber
to interrupted	Light	Plain 40∞watt bulb	0 0	Plain 40∝watt	8 0	Plain 200-watt bulb	Plain 40-watt bulb	Plain 40 cm att bulb	C O	Colored 60-watt bulb	Colored 100-watt bulb	Colored 60-watt bulb	Colored 60-watt bulb
, 1	Experi- ment_no。	Ч	Control	Ŋ	Control	ω.	4	S.	Control	9	2	to	6

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Table 5.--Series iv--Reactions of tuna to continuous radiated white and colored light and

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Throughout the series there was no definite positive tropism to radiating white light from a 40-watt (47 foot candles) bulb with either continuous or interrupted light. Although in Experiment 1 there were 2 entrances, in Experiments 2, 4, and 5 there were none. With continuous light from green, red, amber, and yellow bulbs of intensities ranging from 6 to 62 foot candles, there were no entrances into the field, even though the 100-watt yellow "insect repellent" light had an intensity (62 foot candles) only slightly less than that of the 60-watt white bulb (70 foot candles) which attracted the fish in Experiment 3 of Series iii (table 4).

Lack of response may have been due to the low intensities used, to a lack of responsive condition in the fish, or to a combination of both. Lack of responsive condition is indicated in Experiment 3 using continuous white light from 200-watt bulb in which only 3 entrances were made as compared with 11, in Experiment 3, Series ii (table 3). There was no apparent reason for the difference in behavior. As all of the experiments were low intensity sources except one (Experiment 1), and also the controls, produced 0 entrances, it is believed that the lack of response was due partly at least to the low intensity of the various white and colored bulbs.

Series v-Reactions to the horizontal beam of a projector and to radiating white and colored light from bulbs.

These experiments were performed to study the reaction of tuna to a strong (530 foot candles) beam of white light, and to repeat and extend the observations of Series iv on the reaction to colored light of low intensity. One yellowfin and five tunny were present. The results are shown in table 6_{\circ}

The experiments were conducted with the projection lantern. In Experiment 1 there was one entrance by the yellowfin, but on four occasions the mixed school approached the border of the field and then turned back. In Experiment 2 there was no reaction, the fish behaving as under control conditions. That the fish were in a moderately responsive mood is shown by the 6 entrances under stimulation of a 200-watt bulb in Experiment 3. It would seem that the tuna were not attracted, to the field at least, by the strong continuous light beam.

In experiments 4 to 6, light bulbs fitted with red, green, and blue filters were used, giving intensities somewhat higher than those of the bulbs of comparable color used in Series iv_{0} but less than those of the arc lamp beam of comparable color used in Series i. The green light appeared to attract the tuna, but it is doubtful if the results are of general significance. Neither red nor blue light seemed positively attractive.

SOTT	Remarks	Entrance by yellowfin; others turned back 4 times at a short distance from border			Entered in schools		Entrances by yellowfing tun-	ny schools turned back at border												
EUL LTOW U	Number of entrances to fleld in 5 min.	Ч	0	0	6	0	Ś		0	0	0	0	0	0	0	0	0		m	0
TT nen BTNRI	Intensity (foot- candles)	530	530	0	215	36	28		6	0 0	0°4	4°8	2°8	0°3	0°1	8 Q	530		215	C ð
naur cornier	Rate of interruption per minute	8 0	9 Ð	0	0	0	0		0	0	0	0	0 0	0	0	0 0	70		30	0
an THM Sh	Color	white	white	0	white	red	green		blue	0	red	yellow	amber			0 8	white		white	0 8
SOTING MOLIFICITIES OF THE STORE AND	Light	500=watt projector beam	500-watt projector beam	0	Plain 200-watt bulb	Bulb with filter	Bulb with filter		Bulb with filter	Û 0	Colored 60-watt bulb	Colored 100-watt bulb	Colored 60-watt bulb	Colored 60-watt bulb	Colored 60-watt bulb	0	500=watt projector	beam	Plain 200-watt bulb	0 0
	Experi- ment no.	Ч	2	Control	m	4	Ś	51	6	Control	2	80	6	10	11	Control	12		13	Control

Table 6.--Series v--Reactions of tuna to continuous and interrupted light from projector and to continuous white and colored radiated light from bulbs

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In Experiments 7 to 11, colored bulbs were used, the intensities of which were reduced to 0.1-4.8 foot candles by placing a resistance in the circuit. No attraction whatever was noted at these low intensities.

In Experiments 12 and 13, the reaction of the fish to intermittent light was compared at high and medium intensities. With 70 interruptions (on and off periods equal) per minute, 530 foot candles, there was no positive response. With 30 interruptions per minute (on and off periods equal), the tuna entered the field 3 times, whereas under control conditions it did not enter at all.

Series vi--Reactions to white and colored beams of light from a projector

As the results of the experiments in Series v suggested that light over 500 or below 50 foot candles is ineffective in attracting the tuna, the present series was conducted with white and colored light of moderate intensity (70 to 324 foot candles) from the projector lantern. The intensity of the beam was reduced by placing a resistance in the circuit. One yellowfin and five tunny were present. The results are shown in table 7. As indicated by the relatively large number of entrances (6, 8, and 5) under control conditions, the fish were more active and restless than usual for some unknown reason.

In Experiments 1 and 2, using continuous white light, positive tropism was observed, as indicated by 9 and 14 entrances compared with 6 under control conditions.

In Experiments 3 to 6, colored filters were placed across the projector beam. Although Experiments 3 and 4 were not conclusive, there seemed to be a definite tropistic reaction in the duplicate Experiments 5 and 6 with red and green filters, as shown by 17 and 10 entrances as compared with 8 under control conditions.

In Experiment ?, white light from the projector (324 foot candles) was interrupted $7\frac{1}{2}$ times per minute (equal on and off periods). The tuna entered the field 3 times, and were turned away at the border 14 times when the light changed. In Experiment 8 with white light from a 200-watt bulb, similarly interrupted, the tuna entered the field 8 times as compared with 5 times under control conditions.

llight from 500=watt	Remarks	Positive tropistic reaction		Positive tropistic reaction	Entrance 2 times inside field and 6 times at border	Entrance 9 times inside field and 4 times at border	Tropistic reaction	Tropistic reaction	High number of entrances	Entrance 3 times inside field; turned away 14 times at border	Scared reaction	
n terr upteó	Number of entrances to field <u>in 5 min</u> .	14	9	6	t 0	С Т	17	10	80	m	100	Ś
iusuas and i	Intensity (foot- candles)	324	0 0	324	64	70	64	70	0 0	324	215	0 0
tuna to contir	Rate of interruption per minute	6 0	0 0	0 0	0 9	Û B	0 C	û 0	D D	t.	75	0 0
ction of	Color	white	0 0	white	red	green	red	green	0	whîte	white	0
Table 7Series viReaction of tung to continueds and intermupted light from 500-watt projector.	Light	Projector with resist-	ance	Projector with resista ance	Projector with filter	Projector with filter	Projector with filter	Projector with filter	0 8	Projector with resistance	Plain 200-watt bulb	C D
F	Experi- ment no.	Ч	Control	Ŋ	Ś	4	Ś	6	Control	6	€0	Control

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DISCUSSION

It should be emphasized that the experiments which have been conducted were exploratory in nature, and lead only to tentative conclusions. If some of these appear to have practical application they should be checked by further studies conducted according to planned experimental designs, and with suitable statistical analysis.

It should also be emphasized that the strength of the response to light stimuli varied to some extent from night to night (apart from obvious extraneous sources of interference) indicating a variation in the responsive condition of the fish. Therefore the results of individual experiments are not strictly comparable from series to series.

It should further be emphasized that the experiments were conducted in a relatively small tank and hence under highly artificial conditions. Although certain suggestions may be made as to the response of tuna in the open sea to light stimuli, these should be accepted with great caution.

Reactions to continuous white light

The reactions of the tuna to continuous white lights are summarized in table 8, omitting only those experiments which were disturbed by known extraneous factors. From the many experiments which showed positive results, it is evident that the tuna undergo a change in behavior when exposed to a continuous white light of source intensity between 70 and 450 foot candles. This change in behavior is judged to be a tropistic response to the stimulus, in that the fish approached closer to the source under experimental conditions as compared with control conditions. Morecver, with a horizontal beam, as opposed to light radiated in all directions from a bulb, they approached the source in a line parallel to the beam and turned away only after entering the field and approaching the baffle. There was no conclusive difference in the strength of the reaction between intensities of 70 and about 450 foot candles, although apart from the results with the arc lamp (the higher estimated intensity) there seems to be a tendency for greatest response (experimental minus control entrances) at an intensity of 106 foot candles. On the other hand, weak or no response was obtained with light of low intensity (47 foct candles) and also with light of high intensity (530 foot candles).

It seems possible that tuna could be attracted to a light at sea. However, this would have to be a very high intensity to penetrate the water for any great distance. Although the tuna might be attracted to a light of high intensity, it is doubtful if they would approach very close to the source. Table 8.--Summary of reaction to continuous white light of different intensities

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Experiment and series number	IV = 1 IV - 2	III - 3	11 - 4 111 - 1 111 - 2 111 - 4	Η Η Η Η Η Η Η Η Η Η Η Η Η Η Η Η Η Η Η	VI - 1. VI - 2	ц ц С ц С ц	V = 1 V = 2
ntrances <u>Control</u>	00	Ś	ч 0 %	4400	60	<i>ო ო</i>	00
Number of entrances Experiment Control	20	13	អភ ្ ភភូមិ	4000	14 9	14 18	40
Intensity (foot-candles)	47	02	106	215	324	450 (1)	530

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Reactions to intermittent white light

The response of the tuna to intermittent light of various intensities and rates of interruption are summarized in table 9. At low and high intensities (47 and 530 foot candles) there was no positive response at any rate of interruption which was tried. At an intensity of 106 foot candles, there was positive response but there was no clear relationship between its strength and the rate of interruption. There is, perhaps, a suggestion that the slower rates produced stronger reaction, but this cannot be proven with the present data. Certainly, however, the reactions with interrupted light were no more pronounced than with continuous light. At 215 and 324 foot candles, the results were erratic. Although the fish were attracted towards the source, they were scared away at or near the border of the field when the light was switched on or off. This "scared reaction" was not so pronounced with the weaker light.

The results indicate that in attempting to attract tuna at sea, interrupted white light would give no better response than continuous white light, and moreover, that the fish would probably approach less closely to the source.

Reactions to continuous colored light

With one exception (Experiment 5, Series v), continuous colored lights of intensity of about 4C foot-candles or less, did not induce a positive reaction from the tuna. This was also the case with a yellow "insect repellent" light of somewhat higher intensity (62 foot candles).

On the other hand, continuous colored lights of higher intensity (about 7C foot candles) evoked about the same response as continuous white light of moderate intensity (70 to about 45C foot candles). The response was evidently to the intensity rather than to the color. There was no evidence that light of any one color was a stronger stimulus than a light of any other color for lights of approximately the same intensity.

There is no indication that colored lights of the wave lengths which were used would be of any advantage over white light in attempting to attract tuna at sea.

rescuron to intermittion? White light of different intensities and finterruption	Experiment and number series	IV = 5 IV = 4	111 111 111 111 111 111 111 111 111 11	VI = 8 V = 13 II = 7 II = 10	77 - 7V	γ ~ 12
te light of d	entrances <u>Control</u>	00	0441040	N044	х¢	0
TUM QUALLENSE	Number of entrances Experiment Contro	00	9119 <i>2</i> 9	∞ <i>∾</i> 0.4	n	0
0	Rate of interruption per minute	х 9	6* 60 70 75 100-120	75 75 75	72	70
and rates	Intensity (foot-candles)	47	106	215	324	530

Table 9. -- Summary of reaction to intermittent white light of different intensities and

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*0.5 seconds on, 9.5 seconds off; in all other experiments, on and off pariods were equal.

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PART IV

OBSERVATIONS ON SOUND PRODUCTION AND RESPONSE IN TUNAS/

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Iwac Miyake Associate Professor of Physics University of Hawaii

INTRODUCTION

At the outset of this investigation, two questions were asked, (1) do tuna produce sound, and (2) do tuna respond to sound stimuli? If tuna produce sound, there was the possibility that their presence in the open ocean might be detected by the use of listening devices. If tuna responded to sound stimuli, there was the possibility that the emission of sound of a certain quality and quantity might attract them to a fishing vessel, or that the emission of sound of another quality and quantity might repel them to practical advantage in fishing, e.g., upwards from the lead line of a purse seine during pursing operations.

The exploratory observations reported below were conducted at intervals from August 1951 to January 1952, on tuna confined in a tank and a pond (Tester 1952) at the Hawaii Marine Laboratory, Coconut Island, Oahu, T. H.

MATERIAL AND APPARATUS

When the experiments were first conducted in the concrete tank, it contained but one yellowfin (Neothunnus macropterus) and a few manini (Acanthurus sandvicensis). Later, another yellowfin and a small tunny (Euthynnus yaito) were present. When the experiments were conducted in the large pond, one yellowfin and one tunny were present, along with several small reef fishes of various species.

Three different types of borrowed equipment were used during the course of the work. These consisted of Model CAY Sound Measuring Equipment (reception range, 1 to 10 kilocycles per second) and Model OCP-1 Sonar Test Equipment (reception range 7 to 70 kilocycles per second; transmission range, 5 to 88 kilocycles per second), both loaned through courtesy of local representatives of the U. S. Navy, and NEL Underwater Sound Monitoring Equipment (reception range, about 100 cycles to 10 kilocycles per second), loaned through courtesy of the Navy Electronics Laboratory, San Francisco. In addition, a sound generator consisting essentially of a P-H (Packard-Hewlett) audio oscillator (transmission range, 20 cycles to 30 kilocycles per second) was also used.

^{8/} Contribution No. 25 of the Hawaii Marine Laboratory, University of Hawaii, Honolulu, T. H.

During most of the work the apparatus was installed as indicated in figure 17. In attempting to discover if tuna produced any sound, the hydrophones of either the OAY, OCP-1, or NEL apparatus were suspended 18 inches below the surface of the water near the middle of the tank (about 17 feet from the end), or 17 feet from the inner (unscreened) gate ports at the seaward end of the pond. Sounds in the water were thus detected, amplified, and heard using either earphones or a loud speaker. In attempting to discover if tuna responded to sound stimuli, the transducer of the OCP-1 (a hydrophone used as a transmitter) and of the P-H apparatus were suspended 18 inches below the surface of the water at the south end of the tank, or at the inner gate ports of the pond. The reactions of tuna were noted and recorded under control and experimental conditions.

SOUND PRODUCTION BY TUNA

Procedure

Using one or another of the three hydrophone-amplifier systems, over 100 hours were spent in listening to noises emanating from the tank and pond, and in attempting to ascertain their causes. Because of the possibility of sound production from tuna occurring at some hours and not at others, the observations were spaced to cover all hours of the day and night.

Results

In the tank the hydrophones of the NEL and OAY equipment picked up many sounds, but two were distinctly noticeable over the background noise. One sounded like the snapping of a dry twig and the other was complex, somewhat like that coming from a beaker of violently boiling water.

The snapping sound was identified as that of shrimps. This was established by bringing the hydrophone near a group of shrimps hiding behind one of the corner baffles. The complex sound was for a time thought to be coming from either the tuna or the manini. It was later determined that this sound, and the other background noises, were caused by the water pumping and tank overflow systems.

During one of the night observations, when one yellowfin was present in the tank, three distinctly different sounds were heard. One was a very low frequency sound of very short duration sounding somewhat like a window rattling in the wind. The second sounded like water suddenly stirred with a stick. The third sounded like a stick being dragged over a piece of sheet metal.





FIG. 17. DIAGRAM OF THE TANK (A) AND POND (B) SHOWING THE POSITION OF THE TRANSMITTER AND RECEIVER USED IN EXPERIMENTS WITH SOUND. Later, day and night observations showed that these sounds were caused by tuna in the following manner:

(1) The low frequency sound of very short duration was caused by the sudden movement of the tail of the yellowfin. When getting underway, the first one or two movements of the tail created enough pressure in the water to produce sound waves which were picked up by the hydrophone. Whether this has any biological significance from the point of view schooling is a matter of conjecture. Certainly, in the large pond, the speed undergoes sudden changes while the tuna is swimming its length, and this is also likely to occur in the open sea. It further suggests the possibility that tuna may react to sound of very low frequency.

(2) The second sound was produced when part of the tuna's tail came above the surface of the water. As occasionally tuna are observed to jump and play at the surface, this sound may also be produced in the natural habitat.

(3) The third sound was produced when the tuna accidentally rubbed against the hydrophone.

No additional sounds were detected when two yellowfin and one tunny were present in the tank.

During one of the tests, one of the yellowfin suddenly went beserk, and began bumping and scraping itself against the walls of the tank. When it died, its skin was almost completely eroded. However, even during this period of frantic activity, no sound of moderate or high frequency range (1 to 10 kilocycles) was heard with the hydrophone in use at the time (OAY).

It was decided that a test should be conducted to explore the possibility that tuna might be emitting sound in the supersonic range. A o-hour trial with the OCP-1 apparatus gave negative results. Further trials were not conducted because the apparatus was no longer available.

The NEL equipment was also used to investigate sound production by tuna in the pond. Many different sounds were heard, but none could be identified with either the yellowfin or the tunny.

Summary

Over 100 hours were spent in listening to the sounds picked up by the hydrophone placed in the tank and the pond containing tuna. The listening periods were staggered so that all hours of the day and night were covered. Certain low frequency sounds were produced by the sudden movement of the yellowfin's tail below and at the surface of the water. No sounds emanating from either yellowfin or tunny were detected over a frequency range of 1 to 70 kilocycles per second.

RESPONSE OF TUNA TO SOUND STIMULI

Procedure

It was planned to investigate the reaction of tuna to (1) steady sound of various frequencies, (2) interrupted sound of various frequencies, and (3) complex sounds of short duration. Unfortunately, difficulties with the equipment and the eventual death of the experimental fish, prevented the program from being completed. Such observations as were made pertain to (1), only.

The experiments were started in the concrete tank, and were later removed to the pond. For stimuli over the supersonic range, the OCP-1 equipment was used in the concrete tank. For stimuli at lower frequencies, the P-H equipment was used in both the concrete tank and the pond.

In the latter experiments, the NEL hydrophone was also used at a fixed distance (17 feet) from the transducer. This distance, half the length of the tank, was maintained when the experiments were performed in the pond. The 60-watt amplifier of the P-H equipment was adjusted until the sound became audible at this distance.

To enable any reactions of the tuna to be measured in a roughly quantitative manner, the time which the fish spent in Areas S (sound) and Q (quiet) was recorded under control and experimental conditions. The areas are indicated in figure 17. The sound stimulus was audible (through the hydrophone-amplifier system) in all parts of Area S, but not in Area Q, except near the boundary. It was assumed that the tuna, if attracted or repelled by a sound, would spend relatively more or less time in area S than when there was no sound stimulus.

In the tank, under control conditions, the times spent by the tuna in Areas S and Q should be equal. However, in the pond under control conditions, the time spent in Area Q would be much longer than that spent in Area S because of the relatively greater size of the former area and because, normally, the tuna circles the pond turning at the west end within Area S, and turning at the east end at varying distances within Area Q. Results

7 to 70 kilocycles per second

Experiments at supersonic frequencies were performed in the tank, in which one yellowfin was present. Under control conditions, the yellowfin spent about the same period of time in Area S as in Area Q_{\bullet} .

Supersonic sounds of frequencies of 7_5 10, and in steps of 5 kilocycles thereafter up to 70 kilocycles were generated, each for a period of one-half hour, and the reactions of the yellowfin were observed and recorded. As the results were negative, the data are not included in this report. In each case, the time spent in Area S was about the same as that spent in Area Q.

In these experiments there was no way of knowing whether or not the transducer of the OCP-1 equipment was generating a signal, because at the time, a separate hydrophone was not available. It seems reasonable to assume that it was functioning, as it had been used successfully as a hydrophone in a previous experiment. The results indicate, therefore, that the tuna was not affected by a steady sound of frequency between 7 and 70 kilocycles per second.

500 to 5,000 cycles per second

These experiments were also performed in the tank, using the P-H sound generator and the NEL receiver. Tests, each of 20 minutes duration, were performed using 500_{9} 1,000, 2,000, 3,000, 4,000, and 5,000 cycles. The series was repeated three times. In none of the experiments was there any definite reaction.

To determine whether the yellowfin reacted to a sound frequency other than that used above, the audic oscillator was varied continuously between 100 cycles and 10 kilocycles. There was no noticeable reaction.

100 and 200 cycles per second

As there was the possibility that the tuna in the tank may have become accustomed to strange noises, such as those emanating from the pumps, the apparatus was moved to the large pond where a yellowfin and tunny were present. After a few hours of work, the test came to an abrupt end when saltwater leaked into the transducer, short-circuiting it, and burning out the power output transformer in the amplifier. The fish in the pond died before a new transformer could be procured, thus terminating the experimental work. Such results as were obtained are discussed briefly, as they indicate a possible reaction to sound by the yellowfin. The reactions of the tunny were also observed, but were rarely recorded because of the difficulty of timing two fish at once. The tunny's behavior was independent of that of the yellowfin, and it tended to remain in Area Q.

Under control conditions, with the apparatus in position but not in operation, the yellowfin spent about 6 minutes out of an hour in Area S, passing into the area about 30 times. Successive periods of time spent in Area Q varied from 20 seconds to 3 minutes.

The effect of continuous sound of 100 cycles per second was first tested. The yellowfin spent 10 minutes out of the hour in Area S, entering it 26 times.

During the hour, the noise of the exhaust of a boat was picked up fairly loudly by the hydrophone. Most of the sound doubtless entered the area from the seaward or west end through the screened gates. The fish remained in Area S for a much longer time during this period of disturbance. It also reentered the area after being away for but 20 to 45 seconds.

If the period of disturbance is discounted, the fish remained in Area S for about 6 minutes out of 55 minutes, a result which does not differ greatly from that of control conditions. Although there was no good evidence that the yellowfin reacted to sound of 100 cycles per second, there is the suggestion that it might have been attracted by the complex sound coming from the exhaust of the boat.

The effect of continuous sound of 200 cycles per second was next tested. The tuna spent about 17 out of 47 minutes in Area S, and entered the area 18 times. The results are included in table 10, along with those under control conditions, both to illustrate the reactions, and to show the type of data which were taken.

For some unknown reason, the yellowfin became interested in the hydrophone; it swam up to it six times and appeared to examine it closely. It also circled in the area between the transducer and the seaward end of the pond. Its sudden interest in the hydrophone was peculiar, as it had been swimming past it, without any reaction, for the previous 3 hours. There is the possibility that the yellowfin might have been attracted by a sound generated by the rubber cable rubbing on itself or its support. A strong gusty wind prevailed during this part of the experiment, and although the cable was not observed to move with the wind, it may have done so. On the
other hand, the fish may have been attracted by the 200-cycle sound, and may have been attempting to find its source.

Summary

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The yellowfin in the concrete tank showed no reaction to sound frequencies over a range of 500 cycles to 70 kilocycles per second, nor to a sound with varying frequency from 100 cycles to 10 kilocycles per second.

The yellowfin in the pond seemed to respond to certain sound stimuli, but nothing definite may be stated on the basis of the data now available. Table 10.--Time spent by yellowfin tuna in Areas S and Q in successive circuits of the pond during control (no sound transmission) and experimental (sound transmission at 200 cycles per second) conditions.

Control

Experimental

Circuit	Area S (seconds)	Area Q (seconds)	Circuit	Area S (seconds)	Area Q (seconds)
123456789011234567890122345678903233456	20 25 34 3 30 25 5 2 3 2 5 3 2 3 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 2 5 3 2 5 3 2 5 5 2 2 5 3 2 3 2	$125 \\ 48 \\ 85 \\ 152 \\ 131 \\ 102 \\ 107 \\ 20 \\ 175 \\ 185 \\ 108 \\ 178 \\ 57 \\ 37 \\ 33 \\ 82 \\ 30 \\ 81 \\ 52 \\ 47 \\ 140 \\ 138 \\ 35 \\ 87 \\ 58 \\ 50 \\ 26 \\ 47 \\ 128 \\ 152 \\ 163 \\ 165 \\ 100 \\ 60 \\ 3272 \\ 3272 \\ 3272 \\ 3272 \\ 3272 \\ 337 \\ 338 \\ 300 \\ 81 \\ 510 \\ 100 \\ 60 \\ 3272 \\ 3272 \\ 310 \\ 100 \\$	1 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 6 7 8 9 10 11 2 3 4 5 10 11 2 12 11 12 12	3 30 2 15 300# 180* 20 35* 35 60* 20 2 55* 70* 50* 25 1023	127 200 128 140 122 72 60 30 20 110 80 120 120 120 120 120 103 110 115 45 190

In area between transducer and gates

* At and around hydrophone

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Tester, A. L.

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PART V

NOTES ON THE RESPONSE OF A TROPICAL FISH (KUHLIA SANDVICENSIS) TO INTERRUPTED DIRECT CURRENT?/

by

Albert L. Tester Professor of Zoology University of Hawaii

IN TRODUCTION

A study of the reaction of tuna to electrical stimuli was not attempted for the following reasons: (1) because of the expense of purchasing a generator of sufficient power to produce a reasonably high current density (say, 0.002 amps per square centimeter) in the large (10,663 gallon) concrete tank in which the tuna were kept, (2) because of the danger of harming the tuna, which were being used for other experimental purposes, and mainly (3) because there were indications from the work of Morgan (1951) that further research could be undertaken profitably with the aholehole or "mountain bass" (Kuhlia sandvicensis), using a tank and generator which were already available. Consequently, with the small amount of time that was available, the author attempted to duplicate and extend Morgan's experiments in an effort to discover the optimum pulse duration for minimum power output to attract aholehole in an interrupted, direct current system. It should be pointed out that before seriously considering the practicability of catching tuna or any other fish by electro-fishing methods on the high seas, still more efficient use of the available power than that achieved either by Morgan (1951) or by the Cooperative California Sardine Research Program (anon., 1950) must be made. Otherwise, the power plant required by a fishing vessel would be extremely large, expensive, and therefore probably impracticable.

As the present study represents an extension of Morgan's (1951) work, and as his data are not readily available, his results may be summarized here to advantage. He attempted to discover the minimum current which would lead or force <u>aholehole</u> to the positive pole in a column of salt-water (wooden tank) measuring 12 x 2 x 1 feet, using a source E.M.F. of 220-230 volts, D. C. In some experiments the current was

^{2/} Contribution No. 26 of the Hawaii Marine Laboratory, University of Hawaii, Honolulu, T. H.

used in a continuous flow; in others it was interrupted by a specially-designed interrupter consisting essentially of revolving disks with different proportions of brass and bakelite, the current being "on" when two brushes were in contact with the brass sector, and "off" when the two brushes were in contact with the bakelite sector. Series of experiments were conducted with various currents and with various frequencies of interruption at each current. In other series of experiments the "on-off" ratio was changed, using disks which allowed the current to flow for 0.75, 0.50, and 0.25 of one complete revolution. To maintain a source voltage of 220-230 volts, he found it necessary to design a special type of electrode--a carbon rod enclosed in a plastic tube with open ends and with holes bored through the sides. By adjusting the exposure of the carbon rods to the seawater, it was possible to vary the current in the system, but at the same time to maintain the voltage at the source.

Morgan's results showed (1) that frequency of interruption was not critical for the species and size-range of fish used--about the same response was evoked for frequencies between 15 and 25 r_{oposo} , and (2) that, within the limits of his experiments, the shorter the pulse duration the smaller the average (interrupted) current required to give the desired response -- decisive and rapid movement from the center of the tank to the positive pole. To illustrate, positive response (an entire time period of 60 seconds spent by all fish tested in the half of the tank adjacent to the positive pole) was obtained with an average current of 8 amps and an on-fraction of 0.75, with an average current of 5 amps and an on-fraction of 0.50, and with an average current of 3 to 4 amps and an on-fraction of 0.25. The corresponding current densities may be calculated at 0.0047, 0.0029, and 0.0024-0.0018 amps/cm², respectively. Thus, as the on-fraction of a pulse was reduced, there was a decrease in the average current require to attract the fish and a corresponding saving in power.

In extending Morgan's work, two lines of investigation suggested themselves, (1) to determine the relationship between source voltage and electrode size, and (2) to further decrease the on-fraction to determine the minimal current which would evoke positive response.

SOURCE VOLTAGE AND ELECTRODE SIZE

The wiring hook-up of the apparatus is shown in figure 18. The source of power is an Onan, 2-cylinder, gas-driven, air-cooled, direct current generator unit with a maximum output of 5000 watts (21.8 amps at 230 volts). The source



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FIG. 18 CIRCUIT AND APPARATUS USED IN THE ELECTRICAL EXPERIMENTS.

voltage (maintained by Morgan at 220 volts) may be varied between limits (about 50 to 250 volts) by means of a rheostat in series with the generator field coils. The voltmeter (V_1) measures the source voltage. The voltmeter (V_{0}) measures the voltage after interruption, an average value which is a fraction (approximately the on-fraction) of the source or peak voltage. It also measures the voltage at, but not between, the electrodes if it may be assumed that the resistance of the reversing-polarity switch and the lead wires to the electrodes is negligible. The ammeter (A), which may be placed anywhere in the circuit, measures the average interrupted current (I), which again is a fraction (approximately the on-fraction) of the source or peak current. The interrupter is a motor-driven disk which may be changed to give the desired "on-off" ratio. The electrodes have unknown resistances $(R_1 \text{ and } R_2)$ which, with tube-encased carbon electrodes, vary with the extent of exposure to seawater and the extent of polarizing by gas bubbles which are generated during a current flow. The electrodes are immersed at either end of the tank of seawater. The seawater has a resistance (R3) which may be calculated roughly as

$$R_3 = k \frac{L}{Ar} = 0.0523 \frac{12}{2} = 0.3138 \text{ ohms,}$$

where k is the specific resistance of seawater at a chlorinity of 19 $p_{\circ}p_{\circ}m_{\circ}$ and a temperature of 25^o C_o, L is the length of the water column (feet) and Ar_o is its area (square feet). The apparatus and hook-up described above are identical with those used by Morgan, except for the insertion of the voltmeter (V₂) across the line after interruption.

Of the various voltage measurements which might be made, the average voltage (V_3) across the electrodes is the only one of importance from the point of view of the reaction of the fish. This cannot be measured directly without a special voltmeter, but it may be calculated according to Ohm's Law:

knowing the average current (I) and the resistance of the column of seawater (R_3) . Thus the following values may be obtained:

I (amps)	V ₃ (volts)
1	0.31
2	0.63
3	0.94
4	1.26
5	1.57
etca)

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The average current may be varied by changing the resistance of the electrodes, by varying the source voltage and thus the average interrupted voltage, or by both. Morgan was able to maintain the source (V_1) at 220 volts with increasing average current (I) by decreasing the resistance of the electrodes. This is illustrated by the following empirical determinations which were made with his apparatus (delivering only 210 volts at the time), using tube-encased carbon electrodes, the exposed surface of which was varied, and using an onfraction of 0.25 at 15 r.p.s.:

V _l (volts)	I(amps)	V ₂ (volts)	$R = V_2/I \text{ (ohms)}$
210 210	1 2,5	48 50	48 20
210	<u>4</u>	52	13

The increase in average current was accomplished by decreasing the resistance of the electrodes (increasing their area of exposure), without materially changing either the source or the interrupted voltage. The calculated resistance R is mainly that of the electrodes R_1 and R_2 .

To demonstrate that the same result could be obtained by varying both the source voltage and the resistance of the electrodes, empirical determinations of V_1 and V_2 were made at a constant average current (I) of 3 amps, and therefore at a constant voltage (V_3) across the electrodes of 0.94 volts. The resistance of the electrodes was gradually decreased by changing from small tube-encased carbon rods, to small carbon rods not encased, to large carbon rods wholly immersed, and finally to plates of galvanized iron partially and wholly immersed. In each case, the source voltage (V_1) was adjusted to give an average current (I) of 3 amps with an on-fraction of 0.25 at 15 r.p.s.:

V _l (volts)	V ₂ (volts)	$R = V_2/I \text{ (ohms)}$
210	52	17.3
160	3 8	16
110	21	7
95	16.5	5.5
85	12	4

Since in each of the above determinations, the current (3 amps) and the calculated voltage across the electrodes (0.94 volts) remained the same, the response of fish in the tank should be identical. If low source voltage is a desirable feature, attempts should be made to procure non-

polarizing electrodes of low resistance; in nearly perfect electrodes $R \longrightarrow R_1 + R_2 \longrightarrow 0$. In the present trials, minimum resistance was obtained with galvanized iron electrodes the area of which nearly equalled that of the cross section of the tank. Other types of electrodes were not tried. It may be noted that in the above table, V_2/V_1 progressively departs from an initial 0.25/1 ratio; the reason is not clear.

A series of experiments were planned with a source potential of 95 volts, currents of 2, 3, and 4 amps, and on-fraction 0.25 at 15 r.p.s., to demonstrate that the reaction of the fish was the same as at the higher source voltage (220) used by Morgan. Unfortunately only two fish were available at the time the apparatus was adjusted for this experiment. These were used in replicate trials (reversing the polarity of the electrodes) at 2 amps, but they died before further experiments could be performed due to failure of the aquarium water supply. Of the four trials total positive response was evoked in three and partial positive response on one. Although the data are meager and therefore non-conclusive, they are comparable with those of Morgan at the same current, on-fraction, and frequency but at 220 volts source, and indicate the general validity of the reasoning in the preceding paragraphs.

REDUCTION OF ON-FRACTION OF A CYCLE

Two sets of experiments were tried with a very limited available supply of large <u>aholehole</u>, similar in size (9 to 13 centimeters) to those used by Morgan. In one, Morgan's revolving disk interrupter was used; in the other, a speciallydesigned plunging electrode interrupter was used. The galvanized iron electrodes in the 12 x 2 x 1-foot tank almost covered the cross-section of the water column and had a water-exposed area of about 1700 square centimeters. The fish were confined by two gates at the center of the tank. When the current was turned on, both gates were removed simultaneously so that the fish was free to move to either the positive or the negative electrode.

The results are included in table ll. For comparison some of Morgan's results are included in table 12. As total response--rapid and decisive movement to the positive pole from which there is no withdrawal--was desired as the criterion of attraction, both Morgan's and the writer's results are recorded as the number of positive trials and number of non-positive trials, rather than as time spent in the positive half of the tank out of a total test period of one minute.

Revolving Disk Interrupter

By using a half brass, half bakelite disk and staggering the brushes (fig. 19), it was possible to attain an on-fraction of 0.151 (off-fraction, 0.848). The on-fraction was measured as the length of the disk circumference corresponding to the "on" position as determined with an open-closed circuit indicator (ohnmeter), divided by the total disk circumference.

For each of five specimens, two replicate trials (reversing the polarity of the electrodes between trials) were made at an average current of 1.5 amps (Experiment A), and again at an average current of 2 amps (Experiment B) with a frequency in each case of 15.8 r.p.s. (table 11). At 1.5 amps, 8 out of 10 trials were positive. At 2 amps, all 10 trials were positive. It may be seen by comparison of the results in table 11 and table 12, that by reducing the on-fraction from 0.25 to 0.151, the current for positive response has been reduced from 4 to 2 amps, with a corresponding reduction in current density from 0.0024 to 0.0012 amps/cm².

It was impossible to further reduce the on-fraction because of the thickness of the brushes. Moreover, even with a relatively weak average current of 2 amps, there was considerable arcing at the make and break, which tended to burn the bakelite and corrode the brass portion of the disk. Further experiments with the disk-type interrupter were not undertaken.

Plunging-electrode Interrupter

To attain finer adjustment of the "on-off" ratio and in the hope of reducing the spark at the make and break, a new interrupter was designed (fig. 20). This consisted essentially of a revolving wheel, an eccentrically connected rod, and an electrode which dips momentarily into a glass jar containing oil floating on mercury. The jar sits on a platform which may be raised or lowered, thus permitting fine adjustment of the duration of contact. The wheel was driven at approximately 15 r.p.s.

The apparatus was not satisfactory. It vibrated considerably, causing mechanical and electrical connections to break, and more serious, causing irregular waves on the surface of the mercury. Despite the use of transformer oil, and later of pure mineral oil, arcing occurred at the make and break. This gradually burned the tip of the electrode, changing the on-fraction. The oil and mercury soon tended to form an "emulsion", which further changed the on-fraction and caused arcing at the surface.





i, 15.8 r.p.s.	
current,	
l direct	8
o interrupted	on-fractions
ponse of aholehole to interrupted direct current, 15	currents and
Table 11 Response of	at various currents and on-fractions.
Table 1	

Number of Trials <u>itive Non-positive</u>	000448	0000 0 0	н <i>w ч</i> ¦ю	-1 ~ ~ ~ ~
Number Positive	๛๛๛๚๏	<u>р</u> ииии	n w d	2 <mark>1 88 %</mark>
Standard Length (cm.)	12.5 10 9.5	10 10 9,5	12°5 13 ° 5 12 ° 5	12.5 13.5 12.5
F1sh Number	エタうよう	エスタイグ	<u> а а е</u>	ч х е
0n- <u>fraction</u>	0°151	0 .151	0°083	0°083
V1 (<u>volts</u>)	8"7	ΟΠ	35 (2)	(1) 09
I (amps)	1.5	2	0°5	Ч
Experi- ment	₽	A	U	A

Condensed from Morgan (1951).	Number of Trials ositive Non-positive	0 - 0 0 0 -	0 0 0 0 0 0	000000
ensed from 1	Number Positive	~ ~ ~ ~ ~ ~ ~ ~	พพพพดอ	0 10 N N N N N D
	Standard Length (cm.)	222 2511 252	12 13 13 13 13 13 13 13 13 13 13 13 13 13	нцц 5. 2. 2. 2. 2. 2. 2. 2. 2. 2. 2. 2. 2. 2.
on-fract:	Fish Number	ユスラルグ	ユスタイグ	エスタイグ
at various currents and on-fractions.	0n= fraction	0°750	0°500	0.250
various c	V ₁ (stlos)	220	220	220
at	I (amps)	∞ -	Ś	4
	Experi- ment	đ	д	o

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<u>_</u> Table 12. - Response of <u>aholehole</u> to interrupted direct current, 15 r.p.s., At modeling answerth and an exceptions from Marrie (10)

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Table 12.--Response of <u>aholehole</u> to interrupted direct current, 15 r.p.s., at various currents and on-fractions. Condensed from Morgan (1951).

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(Contl	

of Trials <u>Non-positive</u>	00000	40000 4
Number Pos i tive	0 0 0 0 0 0	очочир
Standard Length (cm.)	12°5 11°5 11°5	11 11 5, 5, 11 1, 5, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11 1, 5, 11, 12, 5, 12, 12, 12, 12, 12, 12, 12, 12, 12, 12
F is h Number	ユスタイグ	ユタクムグ
0n- fraction	0.250	0.250
V1 (volts)	520	520
I (amps)	ς	R
Experi- ment	9	ы



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FIG. 20. THE "PLUNGING ELECTRODE" MECHANICAL INTERRUPTER.

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Two experiments (C and D of table 11) were performed using fresh mineral oil floating on the mercury, and an on-fraction of 0.083. At 0.5 amps, 12 out of 18 trials with three <u>aholehole</u> were positive. At 1 amp, 21 out of 26 trials (81 percent) were positive. The latter results are comparable with those obtained by Morgan at 3 amps with an on-fraction of 0.25 (table 12-D). Unfortunately in the present experiments there is no guarantee that the on-fraction was maintained exactly at 0.083, nor that the current was maintained exactly at 1.0 amps. However, the results indicate an additional saving of power by further reduction of the on-fraction.

DISCUSSION

The regression of minimal current for total (or neartotal) response on on-fraction is shown in figure 21 for both Morgan's results and those of the writer. The present results, although meager, indicate the continuance of the downward trend. They should be checked with more fish and better equipment, and extended to include still smaller on-fractions.

It may be noted that at 2 amps average current and 15 r.p.s., the on-period is 0.151/15 = 0.01 seconds, or 10 milliseconds. It is interesting to note that according to Houston (1949), Dr. Konrad Kreutzer of Germany apparently used an onperiod of about 2 milliseconds, with a frequency which varied (2 to 20 r.p.s.) depending on the natural wriggling frequency of the particular fish. The present results are therefore in agreement with those of Kreutzer in indicating the desirability of using a short on-fraction. It will be interesting to determine if 2 milliseconds is the optimum value for positive attraction at minimal current. The present results do not agree with those of the Cooperative California Sardine Research program (Anon 1950), in which greater response was found with a longer on-fraction.

It seems useless to conduct further experiments with the present apparatus. Rather, efforts should be made to invent or perfect an interrupter which is rapid in action, free from arcing, and provides an adjustment for varying the "on-off" ratio. This might be electronic or mechanical. Both have been used by Kreutzer (Anon. 1951) but in his work the electronic switch was to be replaced by an impulse switch, developed by Siemens Werke of Germany, which "is quite compact, is completely mechanical, and is said to have an accuracy of 1/100,000 of a second."





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