

cepted with some caution. The differences associated with age in the spinal distribution of the cortical fibers in the present material could be the reflection of differences in the staining characteristics of the degenerating fibers at different ages. This possibility seems difficult to test with present techniques. Therefore, at present and until further data become available, it appears likely that the bulk of the direct corticomotoneuronal connections have not been established during the first days of life. The virtual lack of extremity weakness in the baby monkey (4 days old) after cortical ablation is probably related to this circumstance.

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12. I thank Dr. Mortimer Mishkin, National Institute of Mental Health, and Dr. John M. Atwood, Department of Anesthesiology, University of Maryland School of Medicine, for invaluable help, without which the present study could not have been completed. Support by grant No. B-1131, U.S. Public Health Service.

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7 August 1962

## Imprinting by Force

**Abstract:** An experiment was performed to determine the effect on imprinting strength of forcing the following response during the critical period in chicks. Results of the procedures used indicate that although imprinting occurred with forced following, it was not as strong as it is when the following is voluntary.

It has been established that a wide variety of stimulus objects can elicit the following response, called imprinting, in newly hatched fowls. Hess (1), Moltz (2), and others have investigated many parameters of this process.

For imprinting an animal to an object other than the natural one for its species, a relatively standard technique has been used. The subject is placed

near the object during the critical period and is gradually induced to approach, then follow, the moving stimulus. To my knowledge, in no instance has a comparison been made of the behavior of birds who have "voluntarily" followed an object with the behavior of those who have been forced to make the following response. Such an experiment is reported here. Since, in such an instance, the following itself cannot serve as an indicant of imprinting strength, other responses must be used. In addition to following, several responses are typical of the imprinted chick, such as (i) "distress notes" when the chick is separated from the imprinting stimulus, (ii) "contentment tones," and (iii) huddling under the object and seeking contact with it (1). These responses provide evidence of a bird's having been imprinted to a particular object.

The subjects were 85 Vantress broiler chicks which had been hatched in special isolation compartments in an incubator, then removed to a darkened brooder room in individual ventilated shoe boxes, where they remained until exposed to the experimental procedure. The animals were divided into three groups, one of 31 and two of 27.

The imprinting apparatus consisted of a circular track 12 inches wide with 14-inch vertical walls of masonite. The circumference, measured from the middle of the track, was 20 feet. An aluminum arm extended over the track from a wheel mounted parallel to the floor 22 inches high. The imprinting stimulus, a blue rubber ball 7 inches in diameter, was suspended from the aluminum arm. A small loudspeaker was installed in the ball through which a "peeping" noise was played via a tape recorder. The movement of the ball around the track was controlled by varying the speed of a small motor connected to the wheel by a pulley belt.

The imprinting procedures were carried out when the chicks were between 8 and 28 hours old. The groups were matched with respect to age when exposed to the imprinting procedures. Animals in group 1, the "normal" imprinting group, were taken from the shoe box and placed in the track 1 foot from the blue ball. After 30 seconds the "peeping" noise was initiated in the ball, and at 1 minute the ball was moved away from the chick slightly. The following response was elicited from each chick in group 1 by this method. Animals in group 1 that followed the ball for 100 feet were assumed to have re-

Table 1. Percentages of approach responses to each object with different imprinting procedures.

Procedure	Chicks (N)	Response (%)		
		Ball	Chicken	No choice
Natural imprinting	31	82	5	13
Forced imprinting	27	59.3	7.4	33.3
Nonimprinted control	27	3.7	9.3	87

ceived sufficient imprinting stimulation and hence were removed from the track (1). Animals in group 1 that did not follow for a full 100 feet had at least 90 minutes of association with the ball during the critical period. In either case the chicks were returned to the brooder room at the completion of the procedure, and they remained there in isolation until given the test for imprinting.

Group 2 chicks were treated in the same manner as group 1 chicks until after they were placed in the track near the ball. At this point a flexible collar, connected to the ball by a 12-inch string, was placed around the neck of the chick. The ball was then moved so as to exert a slight forward pressure on the chick. Thirty seconds later the "peeping" was started and the ball was moved slowly forward. The chick was forced to follow for 100 feet. If it fell, the motion of the ball was stopped until the chick regained its feet. At the completion of 100 feet of following the chicks were returned to the shoe box and then to the brooder room. Group 3 was a control group in which the chicks received no imprinting training during the critical period.

The critical period was assumed to have been completed by the end of the third day after hatching. During the fourth day a test of imprinting strength was conducted. The test consisted of placing the chick in the track midway between a live hen and the blue ball. An approach response by the chick was tabulated when physical contact was made with either object. Every chick was given 3 minutes to respond on each of two trials: if it made no approach it was scored "no choice." The position of the hen and ball on the track were alternated on successive trials.

A record was also made when the chick uttered contentment tones, huddled under the ball, and manifested other relevant activity during the test for imprinting.

The results of the preference test,

which appear in Table 1, show that the animals in group 1 made significantly more approaches to the ball than those in group 2 ( $p = .02$ ). In both groups, however, the number of approach responses to the ball was significantly greater ( $p = .01$ ) than chance; this indicates that imprinting occurred in both groups. Where a chick chose the ball on both trials of the test, it uttered contentment tones, hovered under the ball, and made distress calls in an appropriate manner. Responses to the blue ball of group 2 chicks that chose the ball on both trials of the test could not be distinguished from responses of chicks exposed to the normal imprinting procedure. Animals in group 3 exhibited no preference for either stimulus at a significant ( $p = .01$ ) level.

It is concluded, therefore, that imprinting does occur when following is

involuntary. In each case where a chick was pulled by the ball it demonstrated great distress by giving the appropriate call and struggling violently to resist being pulled. Hess's (1) "law of effort" states that imprinting strength is a function of the amount of energy expended in making the following response. In this instance imprinting also occurred when an extreme effort was made to resist following—that is, to move in the direction opposite to that of the imprinting object (3).

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3. This report is a portion of a thesis submitted to the graduate school of Florida State University. The research was carried out under the guidance of Dr. W. N. Kellogg.

9 October 1962

### Alarm Reaction of the Top Smelt, *Atherinops affinis* (Ayres)

**Abstract.** The "alarm" substance of the top smelt, *Atherinops affinis* (Ayres), has been isolated by extraction with methanol or ether from suffocated top smelt. These concentrated extracts, when introduced into an aquarium containing top smelt, induce a strong alarm reaction in the fish, characterized by rapid swimming, jumping, and often severe seizures. The fact that extracts from white surfperch, *Phanerodon furcatus* (Girard); surf smelt, *Hypomesus pretiosus* (Girard); northern anchovy, *Engraulis mordax* (Girard); or Pacific herring, *Clupea pallasii* (Valenciennes) caused only mild excitation in the top smelt indicates the species-specificity of the alarm reaction.

The exceptional sensitivity of the olfactory senses of marine animals is well established (1). Several workers have shown that the ability of salmon to return to their home stream is, at least in part, an olfactory response to differences in organic substances in various streams (2). Others have demonstrated the salmon-repellent action of a substance found in human and sea-lion skin when present at a concentration of 1 part in  $8 \times 10^{10}$  parts of water (3). This substance was later thought to be the simple amino acid L-serine (4).

As a means of warning other members of a school, alarmed fish communicate fright by releasing a chemical substance into the water. This alarm reaction was investigated by von Frisch (5) with minnows. Extracts of minnow skins induced a fright reaction in the

fish; the minnows hovered at the bottom of the aquarium and refused food. Hüttel (6), in investigating the chemical nature of this material, concluded that the "fright" substance is a purine or pterine-like compound.

A recent report by Tester (7) that hungry sharks are more strongly stimulated by a scent released in the water from alarmed prey fish than from undisturbed prey fish has intensified our interest in investigating the alarm substances from saltwater species of fish.

We have found that the top smelt, *Atherinops affinis* (Ayres), exhibits a strong alarm reaction when dilute water solutions of extracts of that species are added to the saltwater aquarium where these fish are living. The alarm reaction consists of rapid swimming back and forth in the aquarium, darting to the surface, occasionally breaking the surface of the water to the extent of flipping out of the aquarium, and finally, making a rapid swimming motion with the mouth against the aquarium glass. The reaction lasts approximately 3 to 5 minutes. Often top smelt will lapse into a seizure, will quiver, and will sink upside down to the bottom of the aquarium. Although they usually recover after 20 to 30 minutes, occasionally these seizures end in death, which may be caused by the fish's striking the sides of the aquarium.

Methanol extracts of the skins or of the whole top smelt (after the fish had

been freshly killed by suffocation) were made, the methanol was removed *in vacuo*, and the semisolid product was dissolved in distilled water for evaluation. After the top smelt had been removed from the methanol they were placed in ether overnight at room temperature, then more of the alarm substance was extracted.

Control experiments with small amounts of methanol and Celite suspended in distilled water to simulate the slight cloudiness of the extracts always yielded negative results. The fish seemed to be unaware of several milliliters of methanol introduced into the aquarium.

There was no response to extracts from top smelt that had been kept chilled for 4 days in ice, or to extracts that had initially evoked a response after these had been stored in distilled water at 0°C for a week. It is suspected that there was bacteriological degradation of the alarm substance. Hüttel (6) mentioned such a problem with extracts from minnows.

After the original strong alarm reaction had been induced and the top smelt had resumed their normal swimming behavior (after 3 to 5 minutes), addition of more of the alarm extract had no observable effect on them. However, the next day a strong alarm reaction could again be evoked by adding the alarm extract.

In order to ascertain the species specificity of the alarm reaction to the top smelt extract, the effects of extracts of other species of saltwater fish on the top smelt and on white surf-

Table 1. Data showing species specificity of alarm substances from saltwater fish. Reactions: (—) feeding; (0) no reaction; (+) mild excitation (quick movements, simulated swimming action with mouth against glass); (++) moderate excitation (rapid swimming or shelter seeking); (+++) strong excitation (rapid swimming accompanied by jumping out of the water).

Extract	Dry weight of extracted material (g)	Reaction	
		Top smelt	White surf- perch
<i>Extracting agent: methanol</i>			
Top smelt	1.18	++ to +++	0
White surfperch	2.70	0	—
Surf smelt	0.94	0	—
Northern anchovy	2.30	+	—
Pacific herring	1.60	+	—
<i>Extracting agent: ether</i>			
Top smelt	0.49	++ to +++	—
White surfperch	0.80	+	—
Surf smelt	6.43	+	—
Northern anchovy	9.70	0	—
Pacific herring	4.30	+	