plicable by the conduction rate of the triggering potential.

The macroflash exhibits rigid coupling to the action potential (*1*). This apparently is also true of the microflash, which, in photometric recordings from individual microsources, occurred in a strictly one-to-one response to each invading action potential. Asynchronous spontaneous flashing of microsources was visually noted in some deteriorating specimens, but never in fresh material. Occasionally, however, large regions of the cell irreversibly failed to luminesce (*7*), and attenuation of the macroflash resulted. This was due, presumably, to failure of invasion of those areas by the action potential, an occurrence occasionally seen in electrical recordings.

Reversible graded alterations in amplitude of the macroflash (that is, summation, potentiation, fatigue) commonly occur as a function of temporal stimulus sequence (*1*). This could occur in at least two ways. (i) Gradations in macroflash intensity could arise from changes in the proportion of sources responding to the triggering potential, and could have their basis in statistical changes in the triggering threshold of the microsource population. This does not, however, appear to be the case, for as noted above, individual luminescent sources flash in a rigid one-to-one response to invading action potentials. Microsources possess little or no triggering lability. (ii) Macroflash gradations could result from summation of gradations in the intensity of the microflashes. This is supported by results of simultaneous recordings of micro- and macroflashes: temporal summation of two closely spaced responses occurs similarly in each. Likewise, macroflash fatigue with repeated stimulation was paralleled by a similar decline in microflash intensity. Thus, the macroflash parallels events at the level of the microsource and can serve as a reliable indicator of microflash kinetics.

Since the triggering potential is all-or-none (*1, 2*), gradations in microflash intensity must result either from changes occurring in the luminescent system itself (for example, as a result of substrate depletion) or at the "control" or "coupling" steps linking the luminescent chemistry to changes in membrane potential. That is to say, summation, potentiation, and fatigue of luminescence must have their physical basis within or adjacent to the microsources.

These data indicate the following. (i) Fluorescent luminescent organelles, 1.5 μ and less in diameter, emit flashes with intensities proportional to their physical size and appear to be the sole origin of light contained in the flash emitted by the organism. (ii) Time courses of microflashes resemble those of macroflashes, but typically show somewhat shorter time constants. (iii) Flashes from different microsources show only minor differences in time course. (iv) Measurable differences in latency between micro- and macroflashes can be explained by the transcellular conduction time (5 to 10 msec) of the action potential. (v) Each source flashes in unison with its immediate neighbors in response to each invading action potential. (vi) Reversible graded changes in intensity of macroflash result from graded changes in amplitudes of microflash. (vii) The kinetics of both flashes are parallel and can be used to investigate fundamental subcellular events in studies on excitation-flash coupling.

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4. A more specific term for these organelles must await electron micrographic clarification of their fine structure. Although there is evidence (R. DeSa and J. W. Hastings, unpublished) that the extracted particulate luminescent systems of *Noctiluca* and *Gonyaulax* exhibit certain physical and chemical similarities, the precise relation between the microsources of *Noctiluca* and the "scintillons" of *Gonyaulax* [R. DeSa, J. W. Hastings, A. E. Vatter, *Science* **141**, 1269 (1963)] cannot be defined at this time.
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9. The photomicrograph is focused in the plane of the thin, complex layer of cytoplasm bounded on the upper surface by the plasma membrane and pellicle, and below by the vacuolar membrane. Dark grey strands are seen in electron micrographs (by Dr. H. S. DiStefano) to consist of cytoplasmic thickenings which include mitochondria, golgi bodies, trichocysts, vacuoles of several kinds, and granules. In the light grey areas between strands, extraordinarily little cytoplasm is found between the plasmalemma and vacuolar membrane, the membranes frequently coming into apparent mutual contact. The dark, round, phase-retarding inclusions have not yet been unequivocally identified in electron micrographs. This photograph fails to convey the slight difference in phase retardation exhibited by microsources (such as paired structures in center of photo) and inactive inclusions.
10. Owing to small movements of the specimen in the plane of the field, the position of the photometer field, as determined photographically prior to each recording, had an estimated uncertainty of 1 μ. Hence, the photometer field in Fig. 3*B* may have included one of the structures closely adjacent to the circle.
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12. I thank P. Specht for building the microphotometer circuit and Miss Francine Brady for culturing the organism. Supported by grants NSF GB-1908 and GB-3566, and PHS B-3664 and NB-05301, and in part by an ONR grant to the Marine Biological Laboratory.

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Enhanced Distress Vocalization through Selective Reinforcement

Abstract. *Eighteen Peking ducklings were imprinted and tested for their tendency to emit distress calls during the presentation and withdrawal of the imprinted stimulus. A subsequent arrangement in which each distress vocalization led to a 5-second presentation of the imprinted stimulus resulted in an enhanced tendency to emit distress calls.*

When young ducklings (like the infants of many species) are separated from their mother, they typically emit a stream of loud distress-like vocalizations. Several researchers have noted that these calls influence the mother in that they increase her tendency to return to the young (*1, 2*). In this study we sought to examine the complement to this process by determining how the duckling's tendency to emit distress calls is influenced by the mother's return. More specifically, we wished to assess the effects of an arrangement whereby the mother appears each time a distress call is emitted.

The problem is complicated by several factors. If the distress call is solely a reflection of emotionality produced by the mother's absence, then the constant availability of the mother by means of a distress call should minimize emotionality and thereby reduce distress calls. If, however, the mother's return serves to strengthen the behavior which precedes it, these arrangements should lead to the reinforcement of distress calls. As a result, the rate of distress calls should increase.

We sought to evaluate these possibilities by imprinting ducklings to a moving stimulus and then arranging that presentation of the imprinted stimulus was contingent upon the emission of distress calls.

The subjects were 18 Peking ducklings (*Anas platyrhynchos*) that were hatched in isolation and maintained in individual brooder boxes with continuous access to food and water, and with a 40-watt light bulb for heat.

Two units were used for imprinting and subsequent procedures. These were housed in individual sound-treated and temperature-controlled rooms. Each unit consisted of a plywood box (183 by 76 by 76 cm) divided lengthwise by a fine-mesh brass screen into two approximately equal compartments, one for the duckling and the other for the imprinting stimulus. The imprinting stimulus consisted of a white plastic milk bottle mounted over the superstructure of a model train engine which ran the length of its compartment on HO-gauge track. The lighting was such that when the subject's runway was illuminated and the stimulus compartment darkened, reflections from the screen prevented the subject from viewing the stimulus. When, however, the lighting was reversed, the imprinted stimulus became visible. Stimulus presentation consisted of illuminating the stimulus compartment and moving the stimulus at approximately 30 cm/sec.

Distress calls were detected with specially constructed voice keys which, through selective filtering, were sensitive only to those sounds in the frequency range within which distress vocalizations typically fall (approximately 3000 to 4000 cy/sec) (*3*). A bank of relays, stepping switches, timers, and recorders was employed to program stimulus presentations and to document distress calls.

All subjects were given four imprinting sessions during the first 48 hours after hatch. Each session lasted 45 minutes. The duckling was placed in the runway, the stimulus compartment was illuminated, and the stimulus was moved back and forth along its track for the entire session.

On the two days following imprinting sessions (days 3 and 4), the subject was returned to the apparatus for 40 minutes and during this period the moving stimulus was presented and withdrawn in 1-minute intervals. Thus, each subject was given 20 periods of 1 minute each in which the imprinted stimulus was present and moving, and an equal number of periods in which it was absent. Figure 1 shows a sample record of one of the more proficient ducklings. It illustrates the sizeable performance differences associated with the presence and absence of the imprinted stimulus. This record shows that the subject emits a high rate of distress calls following the removal of the imprinted stimulus and continues to do so until the stimulus is presented. Moreover, the return of the imprinted stimulus is associated with a rather abrupt cessation of distress calls, and relatively few calls are emitted when the stimulus is present.

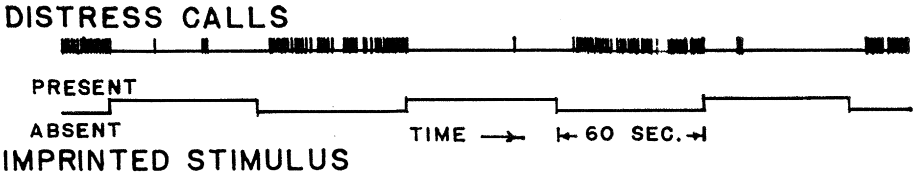

Fig. 1. Sample record of distress calls as influenced by the presentation and withdrawal of the imprinted stimulus.

For all subjects, the mean number of distress calls during stimulus presentation was 210 and 163 on days 3 and 4, respectively, whereas 632 and 485 calls (average numbers) were emitted on these days during the periods when the stimulus was absent. The differences on both days are statistically reliable. Using a test for related measures $t = 5.14$, df $= 17$, $p < .005$ for day 3; and $t = 518$, df $= 17$, $p < .005$ for day 4. These data corroborate the findings of Moltz (*4*) and others (*1*) which indicate that when subjects have been imprinted, the tendency to emit distress calls is determined by the proximity of the imprinted stimulus.

In order to assess individual differences in the strength of the subjects' reactions to the imprinted stimulus, two measures were tabulated. The first, an alarm index, consisted of the number of distress calls emitted by a given subject in the absence of the imprinted stimulus. The second, a relief quotient, consisted of the number of distress calls emitted by that subject in the presence of the imprinted stimulus divided by the sum of that subject's distress calls during presence and absence of the stimulus. Rank order correlations across the two sessions of testing were relatively high ($\rho = +0.65$ and $\rho = +0.73$ for the alarm index and relief quotients, respectively). Thus individual differences in the strength of the subject's reactions to the imprinted stimulus were consistent from one day to the next. In order to assess the degree to which the two indices were related, rank order correlations were calculated for each of the two days. Neither day yielded a significant correlation coefficient ($\rho = 0.10$ on day 3 and 0.02 on day 4).

On the day following the above tests, subjects were paired so as to optimize the match between subjects on both the alarm index and the relief quotient. One member of each pair was then randomly assigned to either the experimental or the control condition. Thereafter, subjects were run with the same partners, and one subject was in each of the isolated units. The programming arrangements for the experimental subject were such that its distress calls immediately produced the moving imprinted stimulus for 5 seconds. The distress calls of its partner had no influence on the presence or absence of the imprinted stimulus. Instead, a yoking procedure was used so that each time the experimental member of the pair emitted a distress call, it produced the imprinted stimulus in its own apparatus and in the apparatus of its partner. This arrange-

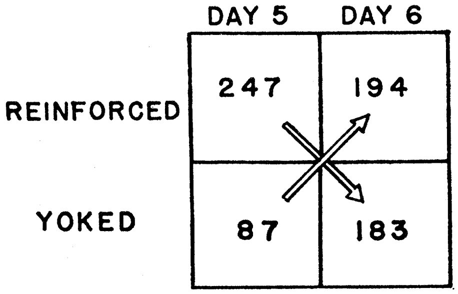

Fig. 2. Mean number of distress calls for reinforced and yoked subjects on days 5 and 6. The arrows indicate the sequence of conditions for subjects within a given group.

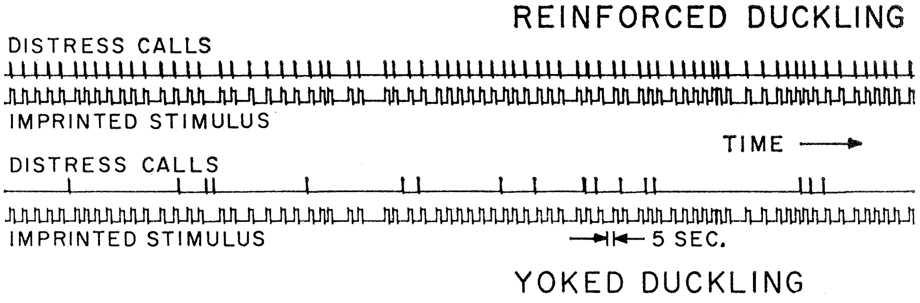

Fig. 3. Sample record illustrating performance differences between the reinforced ducklings and their yoked partners. In this condition presentation of the imprinted stimulus to both ducklings was determined by the distress vocalizations of the reinforced subject.

ment insured that both members of each pair received exactly the same number of stimulus presentations in the same temporal sequence, but, for the experimental subject, presentation of the imprinted stimulus was contingent upon distress calls, whereas for its control (yoked) partner no response contingency existed. On day 5, subjects were run for a 40-minute session and the presence of the imprinted stimulus for both subjects was dependent upon whether or not the experimental subject emitted a distress call. On the following day (day 6), the contingency was reversed for each member of a pair, and now the presence of the imprinted stimulus for both subjects was determined by the emission of distress calls by the previously yoked subject.

Figure 2 shows the mean number of distress calls under each of the several conditions. On day 5, reinforced subjects emitted a greater number of distress calls than their yoked partners. A similar effect is seen on day 6 when the conditions for the members of each pair were reversed, but the observed difference is small.

A test for related measures of the difference in total number of distress calls emitted by the reinforced versus yoked subjects for days 5 and 6 separately yielded a significant value for day 5 ($t = 6.21$, df = 8, $p < .005$) but not for day 6 ($t = 0.16$, df = 8, $p > .05$). In addition, tests for related measures comparing the total number of distress calls emitted by each subject in each of the two conditions show that subjects who were initially yoked (day 5) and subsequently reinforced (day 6) exhibited a significant increase in the number of distress calls emitted ($t = 2.5$, df = 8, $p < .05$). However, subjects going from the reinforced condition to the yoked condition did not show a significant decrease in the number of emitted distress calls ($t = 1.42$, df = 8, $p > .05$).

Figure 3 shows a sample record from day 5. It illustrates the details of the performance difference between reinforced and yoked subjects and provides a basis for interpreting the data in Fig. 2. For the reinforced duckling, the frequency of distress vocalization is relatively high. As a result, the imprinted stimulus seldom is absent for more than a few seconds. When, however, the yoked subject is exposed to the same sequence of stimulus presentations (by means of the distress calls of its reinforced partner) few distress calls are emitted. Apparently, as seen in the performance of the yoked subject (and summarized in Fig. 2), when the imprinted stimulus is presented regularly without regard to subject's vocalization, few distress calls occur (*5*). If, however, stimulus presentations are equated, but stimulus presentation is contingent upon distress calls, the tendency to emit distress calls is enhanced. Since response facilitation of this sort is the expected effect of reinforcement, and since all other factors are equated, it is concluded that distress vocalization is reinforced when the return of the imprinted stimulus is solely determined by the emission of distress calls.

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3. In order to insure that the voice keys would selectively respond to distress calls and not to other sounds, several pilot subjects were placed in the apparatus and repeatedly captured and released. During capture subjects typically emitted a constant stream of distress calls and the voice key consistently responded to them. Movements of the imprinted stimulus, however, and other sounds of the subject (for example, the low-intensity contentment cheep) never activated the voice key.
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5. Figure 1 provides the clue to why short-exposure cycles yield so few distress calls. As seen in Fig. 1, the emission of distress calls does not typically begin until several seconds after stimulus withdrawal. Short cycles of stimulus exposure yield relatively few distress calls because the stimulus often returns before the subject ordinarily begins to vocalize. A process of this sort can explain the large reduction in overall frequency of distress vocalization in going from days 3 and 4 to days 5 and 6. On days 3 and 4 the stimulus was presented and withdrawn in 1-minute cycles, whereas on days 5 and 6 (because of the performance of the reinforced subjects) the average cycle length was only a few seconds.
6. Supported by grant number MH-02433-07 from NIH. We thank Norman Stein, Warren Selekeman, and Phyllis Derderian for their assistance. The participation of J.A. and D.S. was supported by a grant from NSF to Pennsylvania State University for a summer program of research participation for college teachers; Mrs. Derderian's participation was made possible by an NSF grant for undergraduate research.

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Age Differences in Infants' Attention to Patterns of Different Complexities

Abstract. *Infants 3 weeks, 8 weeks, and 14 weeks old were presented with checkerboard patterns which differed in the number of squares they contained. The older the infants, the more complex was the checkerboard that they looked at longest. This result does not appear to depend on changes in visual acuity or accommodation related to age.*

Until recently, results of studies of infants' attention to visual patterns of different complexities seemed remarkably consistent. In spite of the fact that the studies were not directly comparable because they used different stimuli and infants were of different ages, they agreed in finding evidence for the hypothesis that infants prefer to look at more complex rather than less complex patterns (*1*). It is not surprising then to find that this hypothesis has sometimes been used as a *post hoc* explanation for results in other studies. For example, when Stechler