

quency until, on trials 9 and 10, seven of the eight remaining subjects chose the nonshock side (7). Throughout the test trials the latencies of the group given subconvulsive shock were significantly greater than those of the group given convulsive shock. For trial 1, a Mann-Whitney U test indicated a difference in latencies between the two groups significant beyond the .02 level. In the group given convulsive shock the median latency for test trial 1 (7 seconds) was identical to the median latency of the last day of pretraining. The subjects that received subconvulsive shock, however, showed a sharp increase in latencies ($p < .02$, sign test) on the first test trial. These findings indicate that active avoidance of the shock goal box develops with repeated convulsive-shock treatments, but at a rate not nearly as rapid as that found with subconvulsive shock of a much lower intensity. The absence of active avoidance or increase in latency after a single electroconvulsive shock is consistent with previous results (4, 5). In view of the fact that several experiments have shown amnesic effects of a single convulsive-shock treatment (1, 4, 5) our findings suggest that it is highly unlikely that the amnesic effect of electroconvulsive shock is to be explained in terms of its punishing effect.

The amnesic effects of electroconvulsive shock were studied in the second experiment. Subjects were given subconvulsive shock treatments followed by convulsive shock either 5 seconds or 1 hour later. Twenty-one male rats, similar in age and strain to those used in the first experiment but from a different vendor, were first trained, by the same procedures and with the same apparatus as were used in the first experiment. On the first treatment day all subjects were given a 2-ma shock through the pinnae 5 seconds after they were placed in one of the goal boxes. Ten subjects were then given an electroconvulsive shock 5 seconds later. The other eleven subjects were returned to their home cages after the subconvulsive treatment and reintroduced into the same goal box 1 hour later and given a convulsive shock after a 5-second delay. As in the first experiment, treatments and test trials were given on alternate days.

On the first test trial, six of the ten subjects that received an early convulsive shock chose the nonshock side (see Fig. 2). In the group that received a

1-hour-delayed shock eight of the eleven subjects chose the nonshock side. Further, five of these eight subjects that avoided the shock side did so by going to a nonpreferred goal box, while this was true for only one of the six subjects that avoided the shock side in the early-shocked group. The conclusion that the two groups differed on the first test trial is supported by the latencies. The median first trial latency was 26 seconds for the group that received 1-hour-delayed convulsive shock and only 12.5 seconds for the early-shocked group. This difference is significant at the .05 level (Mann-Whitney test). Further, in the group that received 1-hour-delayed shock, the latencies on the first test trial were higher ($p < .04$, 1-tailed sign test) than those of the last pretraining trial. The first test trial latencies of the group that received early shock were slightly, though not significantly, lower than those of the last pretraining trial. This result clearly fails to support the "competing response" interpretation of the effects of electroconvulsive shock. On subsequent trials the latencies of both groups rose markedly and were not significantly different on any day.

The finding that the early-shock treatment did not prevent learning with repeated trials is consistent with other studies that used repeated training trials and convulsive-shock treatments (1).

These studies, considered together, provide further evidence that electroconvulsive shock affects performance both by producing amnesia and by inducing fear, but that the punishing effect of these shocks is considerably less effective than that of less intensive subconvulsive shocks and is found only with repeated treatments. An understanding of the basis of the punishing effect will require additional research. The data provide no support for the view (3) that the amnesic effects of electroconvulsive shock are due to the conditioning of competing responses (8).

JAMES L. MCGAUGH

MILLARD C. MADSEN

Department of Psychology,
University of Oregon, Eugene

References and Notes

1. S. E. Glickman, *Psychol. Bull.* **58**, 218 (1961).
2. E. E. Coons and N. E. Miller, *J. Comp. Physiol. Psychol.* **53**, 524 (1960); M. H. Friedman, *ibid.* **48**, 555 (1953).
3. H. E. Adams and D. J. Lewis, *ibid.* **55**, 302 (1962); D. J. Lewis and H. E. Adams, *Science* **141**, 516 (1963).
4. M. C. Madsen and J. L. McGaugh, *J. Comp. Physiol. Psychol.* **54**, 522 (1961).
5. J. T. Heriot and P. D. Coleman, *ibid.* **55**, 1082 (1962); A. Weissmann, *ibid.* **56**, 806 (1963).
6. W. J. Hudspeth, J. L. McGaugh, C. W. Thomson, *ibid.* **57**, 61 (1964).
7. Two subjects were paraplegic casualties: one after two trials and the other after eight trials.
8. Supported by research grants MY-3541 and MH 07015 from the National Institute of Mental Health.

20 February 1964

Mimicry: Differential Advantage of Color Patterns in the Natural Environment

Abstract. *With a new modification of the release and recapture technique in which day-flying male moths are artificially painted, released, and then lured by their females into traps, it has been possible to obtain differential recapture frequencies in natural areas of the Neotropics.*

During the past 5 years, considerable progress has been made in understanding the evolution of mimicry (1) and experiments with caged animals have conclusively demonstrated that substantial protection from predation by birds, toads, and lizards is gained by mimetic butterflies, flies, and beetles (2). However, measurements of the selective advantage of mimicry actually occurring under natural conditions have not been attempted. We have now been able to make such measurements by means of a novel adaptation of the method of release and recapture de-

veloped by Kettlewell (3). An underlying assumption is that differential recapture is the result of differential predation. This seems justified on the basis of Kettlewell's work.

The new technique takes advantage of several biological properties of the saturniid moth, *Hyalophora (Callosamia) promethea* (Drury), a native of eastern North America. As in all members of this family, the specific sexual attraction of the male to the female is mediated over long distances by a chemical substance known as a pheromone (4). Unlike most saturniids, *H.*

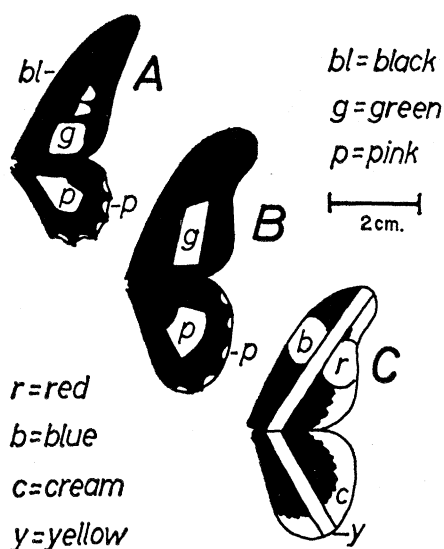


Fig. 1. A, The distasteful butterfly, *Parides neophilus*, slightly enlarged. B, Its experimental mimic, a painted *Hyalophora promethea* male. C, *Hyalophora promethea* male, painted to be uniquely conspicuous.

promethea has two additional properties which we exploited. (i) The courtship flight of the male is diurnal and occurs from approximately 3:30 to 6:30 P.M. (5), so that its period of activity is included within that of diurnal predators. (ii) The males have diverged from the usual coloration of the group and their upper wing surfaces are uniformly black, bordered with cream. On the basis of these two facts Remington (6) has speculated that males of *H. promethea* may mimic the butterfly *Battus philenor* (Linné) with which it is sympatric. That it is palatable was attested by our observations of blue jays, *Cyanocitta cristata* (Linné), eating it in the wild in Massachusetts.

Table 1. Recaptures of experimental and control moths. In experiment 1, experimental moths were painted to mimic *Heliconius erato*, in experiment 2 they were painted to mimic *Parides neophilus*, and in experiment 3 they were painted to be uniquely conspicuous.

Experimental moths		Control moths	
Re- leased	Re- captured	Re- leased	Re- captured
<i>Experiment 1*</i>			
52	16 (31%)	51	18 (35%)
<i>Experiment 2†</i>			
156	45 (29%)	162	44 (27%)
<i>Experiment 3‡</i>			
43	8 (19%)	42	16 (38%)

* Expt. 1: $\chi^2 = .240$, 1 d.f., $.70 > P > .50$.

† Expt. 2: $\chi^2 = .112$, 1 d.f., $.80 > P > .70$.

‡ Expt. 3: $\chi^2 = 3.99$, 1 d.f., $.05 > P > .025$.

Cocoons, obtained commercially, were taken in June 1963 to the field station of the New York Zoological Society in Trinidad. The ecology of this area, which has a rich neotropical biota, is described elsewhere (7). The cocoons were put in a sheltered outdoor emergence cage, and each day newly hatched virgin females were removed to a separate cage. In the evening, the males were colored for the experiments by painting parts of both surfaces of their wings with Flo-Paque paint (8). In order to alter the cream-colored borders of the wings of all moths in experiments 2 and 3 without overburdening them, lightweight marking ink (9) was applied. Black was used in experiment 2, and yellow, which affected the cream color only slightly, was used in experiment 3.

In each experiment two sets of males were prepared: (i) experimentals, painted with Flo-Paque either to resemble a naturally abundant species of distasteful butterfly, or to be brightly colored in a unique manner; and (ii) controls, not changed in color, but painted with comparable amounts of black paint and manipulated to the same extent as the experimentals. All were left overnight in an outdoor cage, and the next morning between 8 and 10 a.m. they were collected and released into the wild. Characteristically they flew to tree foliage and settled, but occasionally continued until out of sight. Approximately equal numbers of control and experimental moths (varying from 1 to 18 of each) were released each day. Although no counts could be made, the wild butterflies vastly outnumbered the moths made to mimic them.

To recapture the males, up to eight virgin females were maintained in each of six assembling traps in the vicinity of the release area. These were plywood boxes 122 cm long by 46 cm square. A conical screen funnel projected toward the inside center from each end of the trap, and between their apices a screened compartment held the females. Thus wind could blow through the trap and spread the female scent, and the males could enter from either end, but could not mate with the females. Separate sliding doors for each of the three compartments permitted removal of the males and addition of new virgins or removal of dead ones. The traps were checked daily in the evening of the same day of release or early the next morning. All moths

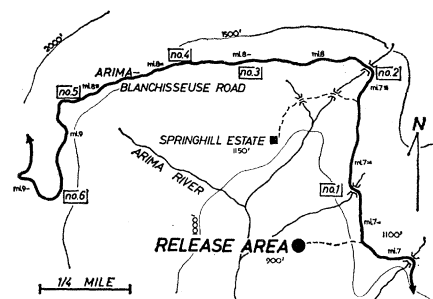


Fig. 2. The Northern Mountain Range, Trinidad, West Indies, where experiments 2 and 3 were conducted. Locations of the six recapture traps are shown.

were numbered on the underside of one wing with small black ink numerals, and recaptured individuals were released again the next morning if still healthy. Out of 506 releases of 406 moths, 313 were released once, 87 twice, 5 three times, and one four times. Recaptures totaled 147 (29 percent) of the 506 releases (10). Since almost all recaptures occurred during the afternoon of the day of release, the possibility of differential mortality by nocturnal predators was eliminated.

The first experiment was carried out on the Waller Field Savanna, a secondary successional area of a deserted World War II air base immediately south of the Northern Mountain Range. The traps were placed around a rectangle of approximately 0.4 by 0.8 km ($\frac{1}{4}$ by $\frac{1}{2}$ mile), and the males were released at a point south of its center. These moths were painted with red oval patches on their forewings to mimic *Heliconius erato hydara* Hewitson, a common species of butterfly in Trinidad which was shown by an earlier study to be highly distasteful to caged birds (2). The earlier study also provided evidence that approximately one-third of the 62 wild-caught birds used had learned to reject *Heliconius* butterflies in their natural environment. It was therefore predicted that a higher proportion of the artificial mimic moths would survive than control, nonmimic moths. However, the frequency of recaptures for the two did not depart significantly from equality (Table 1).

In the second experiment, the moths were painted to mimic another common species of distasteful butterfly, *Parides neophilus parianus* (Huebner) (Fig. 1, A and B), and controls were made uniformly black. These were released at a different site in the Northern Mountain Range. The six traps were placed

at approximately 0.8-km intervals along a semicircle formed by the Arima-Blanchisseuse Road at the head of the Arima Valley (Fig. 2). The point of release was on a hill south of the semicircle and distances to the traps varied from 0.4 to 1.2 km. Recaptures were made at all six traps. The results of this experiment were similar to the first: mimics and control moths, contrary to prediction, were recaptured to nearly the same extent (Table 1).

The failure of these two attempts to demonstrate mimetic advantage is possibly explained by the fact that the artificial mimic and control moths are both relatively inconspicuous and consequently attract such a small amount of predation that differential recapture could not be detected within the magnitude of the experiments.

Therefore, a third experiment was carried out in the same area as experiment 2, with the prediction that a highly conspicuous color pattern that is nonmimetic would be less frequently recaptured than its control. Moths were painted a unique pattern with extremely bright and contrasting colors, but were not painted to resemble any species of Trinidad lepidopteron (Fig. 1C). The frequency of recapture of the painted controls exceeded that of the uniquely painted experimentals by a factor of 2 to 1. This difference is significant at the .05 level (Table 1). The experiment has therefore established that the color pattern of a diurnal insect can influence its survival in the natural environment.

LINCOLN PIERSON BROWER
JANE VAN ZANDT BROWER
F. G. STILES, H. J. CROZE
A. S. HOWER

Biology Laboratory, Amherst
College, Amherst, Massachusetts

References and Notes

1. G. Bernardi, L. P. Brower, J. V. Z. Brower, E. B. Ford, C. S. Holling, D. B. E. Magnus, C. L. Remington, P. M. Sheppard, "Mimicry" (symposium), in *Proceedings, XVI International Congress of Zoology* (XVI Intern. Congr. Zoology, Washington, D.C., 1963), vol. 4.
2. L. P. Brower, J. V. Z. Brower, C. T. Collins, *Zoologica* 48, 65 (1963).
3. H. B. D. Kettlewell, *Heredity* 9, 323 (1955); 10, 287 (1956).
4. P. Karlson and A. Butenandt, *Ann. Rev. Entomol.* 4, 39 (1959).
5. P. Rau and L. Rau, *Trans. Acad. Sci. St. Louis* 26, 81 (1929).
6. J. E. Remington and C. L. Remington, *Yale Sci. Mag.* 32, No. 1 (1957).
7. W. Beebe, *Zoologica* 37, 157 (1952).
8. Floquil Products Inc., Cobleskill, N.Y.
9. Cado Permanent Marker, Esterbrook Pen Co., Camden, N.J.
10. No males of the numerous local saturniid species were trapped over the period of study (9 July to 18 August 1963). This

generates a conflict with Schneider's electrophysiological work in which he found a lack of specificity of saturniid pheromones. Further analysis under natural conditions is greatly needed. [See D. Schneider, *J. Insect Physiol.* 8, 15 (1962); see also E. O. Wilson and W. H. Bossert, *Recent Progr. Hormone Res.* 19, 673 (1963).]

11. Supported by NSF grant 20152 and by an NIH training grant to Amherst College, in conjunction with the New York Zoological Society. We are grateful to E. B. Ford, P. M. Sheppard, H. B. D. Kettlewell, L. M. Cook, W. H. Dowdeswell and J. R. G. Turner for reading and criticizing the manuscript, and to Jocelyn Crane and Miriam Rothschild for valuable comments.

20 January 1964

Estrogen Administered Neonatally Affects Adult Sexual Behavior in Male and Female Rats

Abstract. *The injection of 100 µg of estradiol benzoate into female rats 96 hours after birth abolished sexual receptivity in adulthood, even with estrogen and progesterone replacement after ovariectomy. The administration of testosterone propionate to these animals in adulthood elicited the full pattern of male sexual behavior. The same dose of estrogen given to male rats 96 hours after birth produced adults which were unable to achieve intromission, although they mounted as frequently as normal animals. Testosterone replacement after castration in adulthood reproduced this abnormal behavior.*

There is evidence (1) that secretions of the fetal and neonatal gonads influence sexual differentiation. It has been reported that neonatal female rats receiving a single injection of testosterone propionate in doses of 50 µg or more exhibit constant vaginal cornification and possess atrophic ovaries containing ripe follicles and no corpora lutea. It has also been found that neonatal males given a single injection of estradiol benzoate in doses of 50 µg and above show marked atrophy of the testes.

Although the major interest in the effects of neonatal sex steroids has been on the subsequent reproductive capacity of the adult organism, there has recently been considerable attention (2) directed toward the effects of these neonatal hormone treatments on patterns of adult sexual behavior. Female rats treated neonatally with testosterone propionate in doses of 50 µg or more are sexually unreceptive in adulthood, even when castrated and given estrogen and progesterone replacement. Administration of testosterone propionate to these adult anovulatory females does,

however, elicit male sexual behavior patterns in the presence of a receptive female. Recently Whalen and Nadler (3) reported a study in which neonatal female rats treated with 200 µg of estradiol benzoate are also sexually unreceptive when castrated and given estrogen and progesterone replacement. These data are in contrast to the findings of Harris and Levine (4) that neonatal female rats treated with 50 µg of estradiol benzoate showed sexual receptivity when given estrogen and progesterone replacement and exhibited persistent vaginal cornification with the same ovarian histology as the testosterone-treated females mentioned above. The one obvious difference between these studies is the dose of estradiol benzoate administered to the neonate. It has been shown recently that the dose of the hormone given to the neonatal female is critical with regard to sexual behavior. Barraclough and Gorski (5) have reported that doses of testosterone propionate in the range of 10 µg do not abolish sexual receptivity but, in fact, produce an animal that is constantly sexually receptive.

In order to attempt to reconcile the differences between the data reported by Whalen and Nadler and those found by Harris and Levine, neonatal female rats ($N = 13$) derived from a parent Long-Evans strain were injected with 100 µg of estradiol benzoate in 0.1 ml of oil at 4 days of age. Control animals ($N = 11$) were injected with 0.1 ml of the oil only (sesame oil). All animals were weaned at 21 days of age and weighed once weekly until 105 days of age. When the rats were 65 days old, vaginal smears were taken on all animals for 18 days. At 105 days, the females were tested with sexually vigorous adult males. Approximately 2 weeks after this, all the females were ovariectomized, and at 170 days they were tested with proven males, with no replacement therapy. At 200 days, the animals were given estrogen and progesterone replacement and again tested with proven males, and finally, at 230 days of age, the females were given testosterone replacement and tested for male sexual behavior with a receptive female. With the exception of the final testing for male sexual behavior, all of the test sessions were for a single 15-minute period. The regime used for estrogen and progesterone replacement consisted of injections of 200 µg of estradiol benzoate 72 and 24 hours prior to the test, and an injection of