campal pyramidal neurons appears to be a robust phenomenon, since it has been found among inbred strains of mice (*1*), in rat strains selectively bred for differential two-way avoidance performance (*1*) and, as shown here, in individual mice bred for randomization of their genotype. In all three studies, no other variable within the regio inferior showed such a constant relation to avoidance behavior. In view of the role of the hippocampus in the mediation of two-way avoidance [an intact hippocampus appears to interfere with good performance (*13*)], we hypothesize that the IIP mossy fibers play a role in this behavioral task. Further experiments must show whether this particular projection is directly involved in avoidance performance or whether it merely represents a neuroanatomical "marker" for an undetected correlation, perhaps in the target system—the regio inferior—or in the site of origin—the fascia dentata.

Since both the IIP mossy fiber distribution and two-way avoidance performance co-vary genetically and also after ontogenetic manipulations (*14*), the system that we have studied holds promise for future work on the interaction of genetics and environment in the establishment of neuronal circuitry underlying behavior.

H. SCHWEGLER
H. P. LIPP*
H. VAN DER LOOS
Institute of Anatomy,
University of Lausanne,
1011 Lausanne, Switzerland
W. BUSELMAIER
Institute of Anthropology and Human Genetics, University of Heidelberg,
D-6900 Heidelberg,
German Federal Republic

References and Notes

1. H. Schwegler and H. P. Lipp, *Neurosci. Lett.* **23**, 25 (1981).
2. T. H. Roderick, R. E. Wimer, C. C. Wimer, in *Knowing, Thinking and Believing*, L. Petrinovich and J. L. MacGaugh, Eds. (Plenum, New York, 1976), p. 143; D. Wahlsten, *Brain Res.* **68**, 1 (1974); H. P. Lipp, *Behav. Brain Sci.* **2**, 42 (1979).
3. F. M. S. Haug, *Adv. Anat. Embryol. Cell Biol.* **47**, 1 (1973).
4. S. Laurberg and J. Zimmer, *J. Comp. Neurol.* **190**, 627 (1980).
5. R. P. Barber, J. E. Vaughn, R. E. Wimer, C. C. Wimer, *ibid.* **156**, 417 (1974).
6. W. Buselmaier, S. Geiger, W. Reichert, *Hum. Genet.* **40**, 209 (1978); A. Oliverio, B. E. Eleftheriou, D. W. Bailey, *Physiol. Behav.* **11**, 497 (1973); P. L. Altman and D. Dittmer Katz, *Inbred and Genetically Defined Strains of Laboratory Animals* (Federation of American Societies for Experimental Biology, Bethesda, Md., 1979).
7. H. Anisman, in *The Psychopharmacology of Aversively Motivated Behavior*, H. Anisman and G. Bignami, Eds. (Plenum, New York, 1978), p. 1.
8. Each animal was tested during 5 days in a modified rat shuttle box for 80 to 100 trials per day. Intertrial time was varied randomly between 10 and 20 seconds. The interval between the conditioned stimulus (light) and unconditioned stimulus (shock) was 10 seconds. Electric shock delivery was scrambled (maximum current passed through grid: 100 μA d-c). The shuttle-box scores refer to the percentage of correct avoidance responses observed on day 5. The mice were from an albino strain bred for maximal genetic heterogeneity (Albino/Füllinsdorf, outbred, SPF, stock population about 5000 animals).
9. G. Danscher and J. Zimmer, *Histochemistry* **55**, 27 (1978).
10. Within the rodent hippocampus more IIP mossy fiber terminals are found at its rostral, septal pole than in its more caudal, temporal portions. However, we found (unpublished observations) that quantitative differences in such mossy fiber distribution between inbred strains of mice remain constant at different levels along the septo-hippocampal axis. Thus the differences between individual mice from a defined sampling region are likely to reflect differences in total mossy fiber distribution.
11. Stratum radiatum was defined as Cajal's stratum radiatum minus the suprapyramidal mossy fiber layer. Strata oriens and pyramidale included the IIP mossy fiber projection. Their inclusion or exclusion was irrelevant for our statistical results. The absolute size of the regio inferior refers to the mean surface of a section (grand total/10).
12. R. G. D. Steel and J. H. Torrie, *Principles and Procedures of Statistics* (McGraw-Hill, New York, 1960). Multiple regression analysis was done on HP-85 desk computer (software package Hewlett-Packard 00085-13003).
13. J. O'Keefe and L. Nadel, *The Hippocampus as a Cognitive Map* (Clarendon, Oxford, 1978).
14. Early hyperthyroidism in a rat strain selectively bred for superior two-way avoidance learning (Roman High Avoidance) strongly increases the amount of IIP mossy fiber terminals [J. M. Lauder and E. Mugnaini, *Nature (London)* **268**, 335 (1977)] and drastically reduces the normally excellent acquisition of these animals [H. P. Lipp, H. Schwegler, P. Driscoll, *Neurosci. Lett.* **7** (Suppl.), 46 (1981)].
15. Part of this work has been presented at the 13th annual meeting of the Union of Swiss Societies of Experimental Biology, Lausanne, March 1981 [H. Schwegler and H. P. Lipp, *Experientia* **37**, 14 (1981)]. We thank F. Vogel and P. G. H. Clarke for critically reading the manuscript; J. Beck, M. C. Cruz, M. Gaillard, M. P. A. Grisoni, and S. Methfessel for technical assistance; and C. Vaclavik for typing. The mice were a gift from the Institute of Biological and Medical Research Ltd., Füllinsdorf, Switzerland. This work was supported by the Deutsche Forschungsgemeinschaft (grant Schw-252/2) and by the Swiss National Science Foundation (grant 3.516).

* All correspondence should be addressed to H.P.L.

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Early Experience Determines Song Dialect Responsiveness of Female Sparrows

Abstract. *In a laboratory experiment, female white-crowned sparrows responded almost exclusively to male songs taken from their home dialect region and usually not to songs taken from an alien dialect region. Song dialect populations may represent a level of genetic population structure below that of the subspecies and may play an important role in songbird evolution.*

Male white-crowned sparrows (*Zonotrichia leucophrys*) sing a song of stereotyped form and duration as part of their territorial defense and their behavioral display to attract females (*1, 2*). Each male usually has only one short (~ 2-second) song in his repertoire. The males in a local population share one or more syllables that differ from those of other local populations, resulting in a geographic mosaic of dialects (*3*).

In Colorado, the mountain white-crowned sparrow is a migratory subspecies with distinct dialect populations of various sizes. Our field experience indicates that some dialect populations may number as many as several thousand adults and others only a few hundred or less. Males learn their song during an early auditory-sensitive period; females also learn song but seldom sing (*4*). The usually monogamous male closely attends the female during the few days of egg laying and copulates with her after her solicitation display, in which she tilts her head upward, elevates her tail, squats slightly with wings fluttering, and emits a trilling vocalization (*5*). The posture, reminiscent of lordosis in small mammals, is termed a copulation posture in studies of female brown-headed cowbirds (*6*).

The biological role of the song dialect is of widespread interest (*7*), since a large number of songbird (oscine) species exhibit dialects. The speculation arose early in song dialect research that in some dialect systems a female may use the dialect markers in male song to select a mate from her natal area (*8*). In a field experiment in which tape recordings of songs representing local and alien dialects were played to territorial males, Milligan and Verner (*9*) counted the number of trill vocalizations by females and found that the response to the local dialects tended to be greater than that to the alien dialects, although "only occasionally did female white-crowned sparrows respond to playback during the breeding season." If females mate selectively with males from their natal dialect area, genetic adaptation to local environmental conditions could occur (*10*). The existence of partial barriers to gene exchange among local populations would fit the model of Wright (*11*) which suggests that this population structure is most conducive to rapid evolutionary change.

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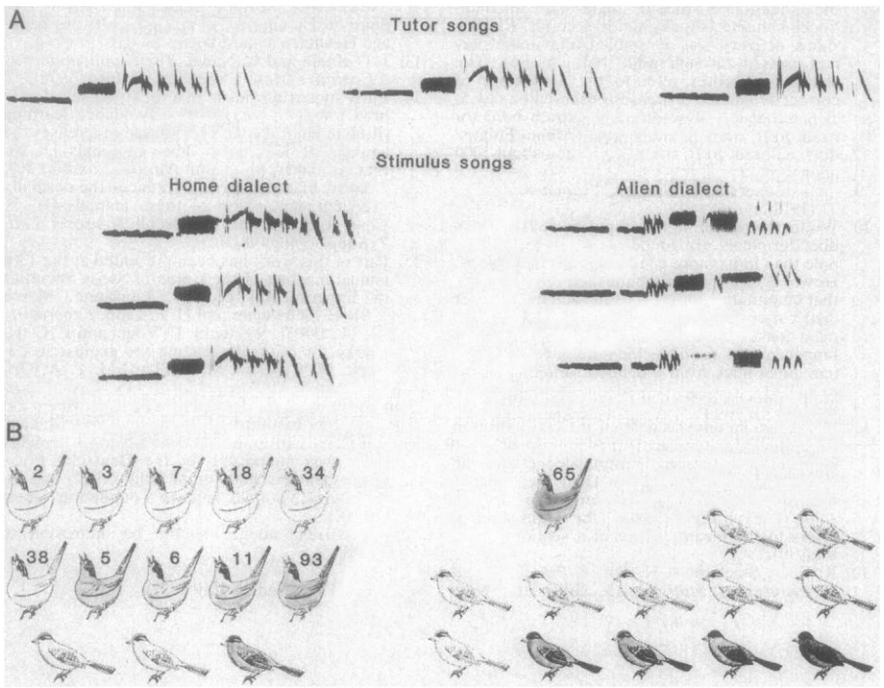

Fig. 1. (A) Sound spectrograms of home-dialect tutor songs and stimulus songs. The spectrograms lie in the frequency range 3 to 6 kHz (vertical axis) and are about 2 seconds long (horizontal axis). (B) Postures showing the response of each female subject to the two stimulus song dialects. Juveniles are unshaded and adults are shaded. The lordotic posture is the copulation solicitation display. Above each female is indicated the number of displays elicited from the subject during a 21-minute test. Juvenile and adult females responded more to the home dialect than to the alien dialect [$P < .05$, $= .07$, and $< .01$ for juveniles, adults, and juveniles plus adults, respectively (two-tailed Wilcoxon matched-pairs signed-ranks test)].

To test the response of females to their natal dialect, we captured nestlings, recent fledglings, and adult females in a montane meadow at Deadman Pass, Colorado (24 July to 4 August). The birds were maintained in the laboratory on a natural photoperiod and exposed for 1.5 hours twice each day to three different male songs, two from the neighborhood where the birds were captured and one from a site 3 km away but in the same dialect (*12*). After the young completed their molt (about 15 September), tutoring was ended and the group was maintained for about 6 weeks on a photoperiod having 8 hours of light and 16 hours of darkness. The photoperiod was then changed to long days, and on 14 to 17 November the birds were implanted with small sections of Silastic tubing containing 17β-estradiol (Sigma); testing was initiated on 24 November. These manipulations with light and hormones induced fully defeathered brood patches as well as sexual behavior.

Tests were conducted in an enclosed chamber, with a single bird in its home cage placed in front of a loudspeaker. Observations were made through one-way glass. A test began with 3 minutes of silence, followed by three repeated segments, each consisting of 3 minutes of stimulus songs and 3 minutes of silence. During each period of stimulation, a subject heard four repetitions of three different songs (12 songs). Thus it was exposed to 36 songs in the 21-minute session (*13*). During each session we tallied the number of copulation solicitation displays and other behavioral acts (*14*). In the experiments reported here each female heard the alien dialect on one day and the home dialect on the next day. Stimulus songs representing the home dialect were recorded from three males at Sand Creek, 9 km from Deadman Pass, and thus were different from the male songs used for tutoring, although of the same dialect. Stimulus songs representing the alien dialect were recorded at Gothic, Colorado, near Crested Butte—about 275 km from the Deadman Pass area.

The response pattern was conclusive (Fig. 1). Juvenile females displayed only in response to the songs of their natal dialect, and adult females behaved similarly. Although one adult female displayed in response to both dialects, she gave significantly more displays to the home dialect (93) than to the alien dialect (65) [$\chi^2(1) = 4.96$, $P < .05$]. By subsequent implantation of testosterone propionate (Sigma), three of the five adult females were induced to sing. Thus we were able to determine that they had learned the home dialect, probably during their early critical period, and therefore were likely to be breeding in their natal dialect area (*14*).

We conclude that female mountain white-crowned sparrows are responsive to male song of their natal dialect and virtually unresponsive to male song of an alien dialect. Early experience with a song sensitized the females to be more responsive to it at a later time. We interpret this response as indicating preference for a sexual partner. Extrapolating to a natural population of mountain white-crowned sparrows, it seems that a male with an alien dialect would be less effective in attracting a mate than would a male singing in the local dialect, since the female would probably discriminate among dialects at the time of pairing. Commonly in passerine populations the sex ratio is skewed in favor of excess males (*15*). Thus we expect that most or all females have a choice of mate on returning to the local breeding habitat from winter quarters and that males with alien dialects are therefore usually less successful in pairing. On the other hand, what might happen to a female who flies off course during spring migration and who had imprinted on a dialect different from the one she now hears at mating time? With an excess of males, she probably would face persistent courtship by bachelor males. Such prolonged exposure might overcome a preference derived from natal experience (*16*). Thus males with an alien dialect may be relatively infrequent in populations, but some mated females may sing in a dialect different from the local one when they are exposed to exogenous testosterone, indicating that they strayed from their natal population. Assuming some resistance to the alien dialect, these females should tend to pair later, on average, than other females.

We suggest that dialect populations are "viscous" with respect to gene flow among the dialects yet have enough leakage of genes to allow the spread of new and possibly advantageous mutations. If so, the rate of evolution would be considerably greater than in a single large panmictic population. Oscine birds do seem to have undergone prolific speciation (*17*).

MYRON CHARLES BAKER
KIMBERLY J. SPITLER-NABORS
DANA C. BRADLEY
Department of Zoology and Entomology, Colorado State University, Fort Collins 80523

References and Notes

1. P. Marler and M. Tamura, *Condor* **64**, 368 (1962).
2. M. C. Baker, *Evolution* **29**, 226 (1975).
3. P. Marler and M. Tamura, *Science* **146**, 1483 (1964); L. F. Baptista and J. R. King, *Condor* **82**, 267 (1980).
4. P. Marler, *J. Comp. Physiol. Psychol.* **71**, 1 (1970); M. Konishi, *Z. Tierpsychol.* **22**, 770 (1965).
5. The female copulation display gradually intensifies over a period of weeks prior to actual copulation. During this period the interest of the male in his mate also gradually intensifies. Posturing and trilling by the female is the immediate antecedent to copulation episodes. Wing fluttering and trilling are evident on other occasions of high arousal in both sexes, but males seldom elevate the tail. A female in a highly aggressive state—as in a fight with another female—displays such masculine behavior as loud song and physical attack, but not sexual posturing. These patterns have been reported in detail by B. D. Blanchard [*Univ. Calif. Berkeley Publ. Zool.* **46**, 1 (1941)].
6. A. P. King and M. J. West, *Science* **195**, 1002 (1977); M. J. West, A. P. King, D. H. Eastzer, J. E. R. Staddon, *J. Comp. Physiol. Psychol.* **93**, 124 (1979); M. J. West, A. P. King, D. H. Eastzer, *Am. Sci.* **69**, 56 (1981); *Anim. Behav.* **29**, 490 (1981).
7. P. Marler, *Ibis* **94**, 458 (1952); R. J. Andrew, *Science* **137**, 585 (1962); F. Nottebohm, *ibid.* **167**, 950 (1970); W. H. Thorpe, in *The New Systematics*, J. Huxley, Ed. (Clarendon, Oxford, 1940).
8. F. Nottebohm, *Condor* **71**, 299 (1969).
9. M. Milligan and J. Verner, *ibid.* **73**, 208 (1971). Audible trill vocalizations and not copulation postures were reported. The data were not presented numerically and there was no statistical analysis.
10. R. B. Payne, in *Natural Selection and Social Behavior: Recent Research and New Theory*, R. D. Alexander and D. W. Tinkle, Eds. (Chiron, New York, 1980). Payne's "genetic adaptation model" is a more rigid statement of forms of the same model discussed by other authors. Electrophoretic evidence from populations of white-crowned sparrows in coastal California [(*2*); M. C. Baker, in preparation] indicates that dialects are genetically differentiated. For a review, see M. C. Baker, in *Evolutionary and Ecological Aspects of Acoustic Communication in Birds*, D. E. Kroodsma and E. H. Miller, Eds. (Academic Press, New York, in press).
11. S. Wright, *Variability Within and Among Natural Populations* (Univ. of Chicago Press, Chicago, 1978); A. H. Miller, *Evolution* **1**, 186 (1947).
12. One subject was a nestling, and the fledglings had been out of the nest for 7 to 10 days when captured. The habitat in which the fledglings were captured is island-like, with the nearest known alien dialect located about 24 km away. Considering their age and geographic isolation, it is safe to assume that the juveniles were locally hatched. The young and adult females were housed individually in Hendrix cages (45 cm long by 23 cm wide by 25 cm high) and fed a diet of mixed finch seed, mealworms, and supplemental vitamins. The birds were tutored with tape-recorded songs played on a Revox A77 recorder through a Realistic SA-2001 amplifier and broadcast with an Altec 755-E speaker mounted in a cabinet. The songs were played at a rate of one each 15 seconds, approximating the natural singing rate of an individual male.
13. The test area was 2 m long by 1 m wide by 3 m high and shrouded with black plastic. It contained an entry door and a table on which rested a 20-cm loudspeaker mounted in a cabinet located 50 cm from a test cage containing an experimental subject.
14. M. C. Baker, K. J. Spitler-Nabors, D. C. Bradley, in preparation.
15. J. L. Brown, *Wilson Bull.* **81**, 293 (1969).
16. Mating between different species often occurs in zoos when normal choice of sexual partners is frustrated.
17. J. A. Feduccia, *The Age of Birds* (Harvard Univ. Press, Cambridge, Mass., 1980); P. Broadkorb, in *Avian Biology*, D. S. Farner and J. R. King, Eds. (Academic Press, New York, 1971), vol. 1; M. C. Baker, *Heredity* **46**, 209 (1981).

18. We thank A. E. M. Baker, L. C. Deutsch, M. A. Cunningham, D. F. Tomback, W. A. Searcy, D. B. Thompson, J. S. Augustine, P. Greco, and G. L. Sherman for assistance and advice. Supported by NSF grant DEB-78-22657 to M.C.B.

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Melatonin: Identification of Sites of Antigonadal Action in Mouse Brain

Abstract. *Long-term implants releasing a small quantity of melatonin (45 nanograms per day) were used to determine the brain sites of the hormone's antigonadal action in a photoperiodic species, the white-footed mouse* (Peromyscus leucopus). *Implants in the medial preoptic and supra- and retrochiasmatic areas elicited complete gonadal regression after 7 weeks. Implants in other brain regions had little effect on the animals' reproductive state.*

The importance of photoperiodism in the control of reproductive cycles has been demonstrated in a variety of mammalian species (*1, 2*). In rodents, photoperiodic control of reproductive state is mediated by the central nervous system through the retinohypothalamic tract, the suprachiasmatic nuclei (SCN), and the superior cervical ganglia (*3*). There is also evidence that the pineal gland and its hormone product melatonin are regulated by these neural pathways (*4*) and that the pineal gland mediates changes in seasonal breeding. In the Syrian hamster and the European vole, pinealectomy renders the animals incapable of responding to a photoperiod with short days, thus preventing regression (*5*). Conversely, daily melatonin injections provoke gonadal regression in the Syrian hamster and the white-footed mouse, *Peromyscus leucopus* (*6*).

Various studies have implicated the ovary, uterus, and anterior pituitary as the sites of melatonin's antigonadal activity (*7*), but there is also evidence for a site in the brain. Melatonin administration induces behavioral and electroencephalographic changes (*8*), and melatonin crystals implanted into the brain reduce the secretion of pituitary luteinizing hormone in castrated rats (*9*). Recently, the hypothalamus was shown to be a putative site of melatonin action. Melatonin is specifically bound in the rat hypothalamus (*10*), and endogenous melatonin has been localized in the SCN by fluorescence immunohistochemistry (*11*).

In a previous study we demonstrated that implants releasing small quantities of melatonin (90 ng/day) were effective in eliciting gonadal regression when implanted in the anterior hypothalamus of *P. leucopus* (*12*). The present study shows that the antigonadal effects of melatonin may be confined to the anterior hypothalamus.

Under chloropent anesthesia (16 mg per gram of body weight; Fort Dodge Laboratories), sexually mature female *P. leucopus* (*13*) were stereotaxically implanted with a melatonin-containing beeswax pellet (*12*) in a predetermined brain region. The pellets released a nearly physiological amount of melatonin (45 ng/day, or 2.4 times that produced by the pineal gland of the Syrian hamster) (*14*). Some mice were implanted with a blank beeswax pellet in the anterior hypothalamus to determine the effects of the implantation procedure on the animals' reproductive state. Correct placement of the pellets was determined histologically at the end of the experiment (*15*). Animals in another group received a subcutaneously implanted pellet of beeswax and melatonin to ascertain whether peripheral release of similar quantities of melatonin has an antigonadal effect. The mice were maintained at 23°C on a photoperiod with 16 hours of light and 8 hours of darkness.

After 7 weeks the mice were killed and the reproductive tract (vagina, uterus, oviducts, and ovaries) was removed and weighed. The mice were also examined for an imperforate vagina, which indicates reproductive regression (*16*). The functional condition of the ovaries was determined by histological examination of the state of follicular development (*17*). The extent of melatonin diffusion from the pellet into the surrounding neural tissue was determined by autoradiography (*18*).

The results indicate that brain sites for the antigonadal action of melatonin are located in the rostral brainstem, notably in supra- and retrochiasmatic areas. Mice with melatonin-containing implants in these regions exhibited a degree of regression of the reproductive tract (Fig. 1 and Table 1) similar to that induced by 12 weeks of exposure to a photoperiod with short days (*2, 19*). Reproductive tract weight decreased 61 percent in mice implanted with pellets in the suprachiasmatic area and 59 percent in mice receiving pellets in the retrochiasmatic area (Fig. 1). A majority of the mice receiving pellets in these areas also exhibited imperforate vaginas and had ovaries that lacked preovulatory follicles (Table 1). Animals with melatonin-containing implants in other brain areas maintained normal reproductive tract weight (Fig. 1). The one exception was a

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