absence of an increase in G1m(3) content in the cerebrospinal fluid of Gm heterozygous MS patients. However, the marked increase of G1m(3) in the cerebrospinal fluid of Gm^3 homozygous MS patients makes this unlikely. Studies of *Gm* gene and haplotype frequencies in MS compared to OND patients and healthy people should provide evidence for or against this hypothesis. Susceptibility to MS is most likely polygenic and might depend not only on genes for histocompatibility antigens (*HLA*) (*21*), but also on *Gm* genes, which are on a different chromosome (*22*). In man, immune responsiveness to flagellin (*23*) and susceptibility to autoimmune chronic active hepatitis (*24*) involve interaction between histocompatibility antigens and Gm. Non–antigen-specific binding of IgG molecules to cell structures such as Fcγ receptors could occur in MS (*9*). Preferential interaction between Fcγ receptors and a given immunoglobulin subclass (*25*) or G1m allotype has been observed (*26*), and our finding of increased levels of G1m(1) (located on the Fc portion of IgG1) may indicate a role of such interactions in the demyelination that characterizes MS.

JEAN-PHILIPPE SALIER
JEAN-MICHEL GOUST
JANARDAN P. PANDEY
H. HUGH FUDENBERG

Department of Basic and Clinical Immunology and Microbiology, Medical University of South Carolina, Charleston 29425

References and Notes

1. J. B. Natvig and H. G. Kunkel, *Adv. Immunol.* **16**, 1 (1973).
2. World Health Organization, *Eur. J. Immunol.* **6**, 599 (1976).
3. S. D. Litwin and S. Balaban, *J. Immunol.* **108**, 991 (1972).
4. J. P. Salier, H. Sarvas, H. M. Reisner, A. C. Wang, H. H. Fudenberg, *Mol. Immunol.* **16**, 217 (1979).
5. H. Link, E. Möller, R. Müller, E. Norrby, J. E. Olsson, L. Stendhal, G. Tibbling, *Acta Neurol. Scand. Suppl.* **63**, 173 (1977).
6. B. Vandvik, J. B. Natvig, D. Wiger, *Scand. J. Immunol.* **5**, 427 (1976).
7. S. Al Kassab, L. Dittmann, H. Olesen, *Acta Neurol. Scand.* **60**, 129 (1979).
8. W. W. Tourtellotte and B. I. Ma, *Neurology* **28** (Suppl.), 76 (1978).
9. D. H. Mattson, R. P. Roos, B. G. W. Arnason, *Nature (London)* **287**, 335 (1980).
10. E. Norrby and B. Vandvik, in *Immunopathology*, P. A. Miescher, Ed. (Grune & Stratton, New York, 1976), pp. 161–172.
11. G. N. Vyas, H. H. Fudenberg, H. M. Pretty, E. R. Gold, *J. Immunol.* **100**, 274 (1968).
12. J. P. Salier *et al.*, *J. Immunogenet.* **7**, 123 (1980).
13. H. Link and O. Zettervall, *Clin. Exp. Immunol.* **6**, 435 (1970).
14. H. Iwashita, F. Grunwald, H. Bauer, *J. Neurol.* **207**, 45 (1974).
15. F. Bollengier, N. Rabinovitch, A. Lowenthal, *J. Clin. Chem. Clin. Biochem.* **16**, 165 (1978).
16. J. P. Antel, B. G. W. Arnason, M. E. Medof, *Ann. Neurol.* **5**, 338 (1979).
17. E. L. Reinherz, H. L. Weiner, S. L. Hauser, J. A. Cohen, J. A. Distaso, S. F. Schlossman, *N. Engl. J. Med.* **303**, 125 (1980).
18. J. M. Goust, P. M. Hoffman, J. Pryjma, E. L. Hogan, H. H. Fudenberg, *Ann. Neurol.* **8**, 526 (1980).
19. C. C. Chien and J. S. Ingraham, *J. Immunol.* **118**, 1896 (1977).
20. J. P. Salier, C. Rivat, L. Rivat, C. Bastard, G. Virella, H. H. Fudenberg, A. C. Wang, *Mol. Immunol.* **17**, 229 (1980).
21. J. L. Tiwari, S. E. Hodge, P. I. Terasaki, M. A. Spence, *Am. J. Hum. Genet.* **32**, 103 (1980).
22. C. M. Croce, M. Shander, J. Martinis, L. Cicurel, G. G. D'Ancona, T. W. Dolby, H. Koprowski, *Proc. Natl. Acad. Sci. U.S.A.* **76**, 3416 (1979).
23. S. Whittingham, J. D. Mathews, M. S. Schanfield, J. V. Mathews, B. D. Tait, P. J. Morris, I. R. Mackay, *Clin. Exp. Immunol.* **40**, 8 (1980).
24. S. Whittingham, J. D. Mathews, M. S. Schanfield, D. B. Tait, I. R. Mackay, *ibid.* **43**, 80 (1981).
25. D. Gilbert, M. Daveau, C. Fontaine, *Scand. J. Immunol.*, in press.
26. M. Daveau, unpublished data.
27. We thank W. W. Tourtellotte, Wadsworth VA Hospital, Los Angeles, Calif., for supplying coded samples from the Human Neurospecimens Bank, M. Daveau, J. P. Raoult, and R. Sesboué for fruitful discussions, and C. L. Smith for editorial assistance. Publication No. 437 from the Department of Basic and Clinical Immunology and Microbiology, Medical University of South Carolina. Research supported in part by PHS grants CA-25746 and NS-16269 and by National Multiple Sclerosis Society grant RG-1167-B2.

22 December 1980; revised 9 March 1981

Development of Stereopsis and Cortical Binocularity in Human Infants: Electrophysiological Evidence

Abstract. *Dynamic random-dot stereograms and correlograms were used to elicit visually evoked brain potentials from human infants, and these potentials were compared with potentials evoked by classical checkerboard pattern reversal. The results indicate that infants begin to produce stereoscopically evoked potentials at the age of 10 to 19 weeks, several weeks after showing classical checkerboard-evoked potentials, and suggest that the onset of cortical binocularity precedes stereopsis.*

There has been increasing interest in assessing binocular visual function in human infants, since early detection of defects could aid in the treatment of strabismus and in the prevention of amblyopia and loss of depth perception. Behavioral responses to random-dot stereograms have been used to demonstrate stereoscopic discrimination in infants 3 to 6 months of age (*1*). An electrophysiological method (*2, 3*) has revealed that infants as young as 2 months can detect changes in binocular spatial correlation between random-dot patterns presented to both eyes (*4*). We report electrophysiological evidence indicating that the visual system begins to process random-dot stereograms when the infant is 10 to 19 weeks of age, several weeks after the processing of luminance patterns begins.

Dynamic random-dot correlograms and dynamic random-dot stereograms contain stereoscopic cues that can only be perceived by subjects with functional stereopsis (*5–7*). When viewed monocularly or by subjects without stereopsis, these correlograms and stereograms appear similar to the "snowstorm" on an untuned television set. Dynamic random-dot correlograms and stereograms, unlike static ones, are devoid of monocular cues caused by binocular correlation changes or by binocular disparity changes (*8*). This is important in measuring visually evoked potentials (VEP's), where repeated presentation of the same stimulus and averaging of the stimulus-locked brain potentials are required.

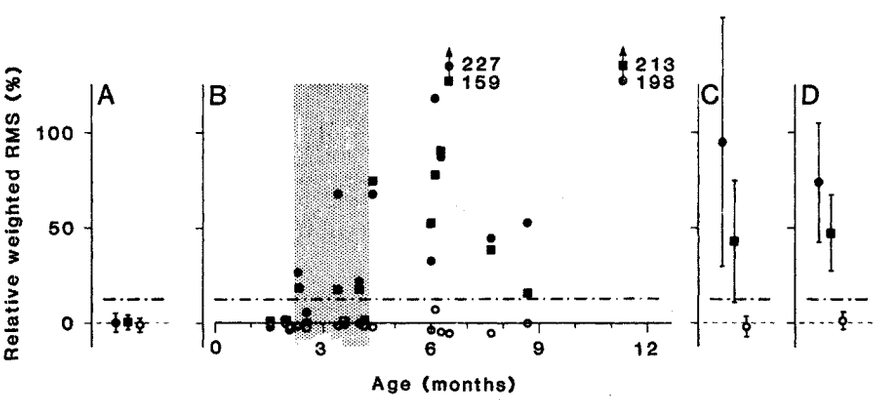

Fig. 1. Relative weighted root-mean-square (RMS) values of the two stereoscopic test stimuli NP (●) and DF (■) and of the control stimulus SS (○). Data are given for (A) stereoblind adults and children (N = 15), (B) infants (N = 17), (C) stereonormal children (N = 20), and (D) stereonormal adults (N = 20). Criterion responses (relative weighted RMS ≥ 12 percent) are above the dashed line. The shaded zone (2 to 4.3 months) is the age range where cyclopean VEP's developed in our sample of infants. Data in (A), (C), and (D) represent means ± 1 standard deviation.

0036-8075/81/0918-1402$01.00/0

We used four types of visual stimuli, each presented stereoscopically to the subjects. Two (the correlogram and cyclopean checkerboard) were test stimuli and had stereoscopic cues, and two (the snowstorm and black-and-white checkerboard) were control stimuli and lacked stereoscopic cues. Each stimulus consisted of two parts that were presented alternately for 200 msec.

Correlogram (NP). A dynamic random-dot pattern was presented to one eye and its inverse to the other eye during the first half of the stimulus period, N (negatively correlated two-dimensional patterns). An identical dot pattern was presented to both eyes during the second half, P (positively correlated patterns). In subjects with normal stereopsis, the correlogram stimulus gave rise to a percept of depth (but with no distinct depth planes or stereoscopic contours) described as "wooly" (N) that alternate with a flat, zero-depth plane of random dots (P).

Cyclopean checkerboard (DF). In the first half of the stimulus, D, the binocularly disparate areas of a dynamic random-dot stereogram were arranged as "squares" which hovered in front of neighboring squares like the white squares in a checkerboard, but without monocular cues. The squares subtended 280 by 280 minutes of arc (14 by 16 dots) and had a binocular disparity of 40 minutes of arc (2 dots). Part D was alternated with the second half, part F, a flat random-dot plane with no binocular disparity. Subjects with normal stereopsis perceived a cyclopean checkerboard jump forward in depth (D) from a flat plane (F) and then collapse back into it.

Snowstorm (SS). This control stimulus was generated in the same manner as stimulus DF except that the pattern that had been presented to one of the eyes in DF was here presented to both eyes. Stimulus SS imitated monocular viewing of DF and resulted in the absence of the pulsating cyclopean checkerboard observed in DF, since both eyes were stimulated identically. The VEP to SS, therefore, was a baseline measurement to ensure that no stimulus-locked artifacts were produced by the experimental equipment. Stereoblind adult subjects observed no perceptual differences between the stimuli NP, DF, and SS (*3*).

Black-and-white (classical) checkerboard (BW). Individual black or white squares, subtending 280 by 280 minutes of arc, were alternated with the opposite color. Both normal and stereoblind subjects could perceive this stimulus. The resulting VEP served as a reference to which the VEP's elicited by the other three stimuli were compared.

The VEP's were recorded from the scalp of infants (*9*), who were held by a parent. Fifty successive VEP segments of 800 msec (two stimulus periods), time-locked with the stimulus, were measured (*10*), corresponding to a total of 100 consecutive stimuli. The VEP segments were averaged during stimulation, and the average VEP was stored for analysis. In general, four VEP's were obtained for each stimulus (in the sequence BW, NP, DF, SS, BW, and so on) for a total of 16 measurements per session. After each session, which lasted approximately 30 minutes, the mean VEP's of the four measurements for identical stimuli were computed.

A weighted root-mean-square (RMS) value was calculated from each mean VEP (*11*). The weighted RMS values for stimuli NP, DF, and SS were each divided by the weighted RMS value for stimulus BW to yield a relative, weighted RMS value for each of the three stimuli (*12*). Control studies in children older than 3 years and in adults demonstrated that in all subjects with stereopsis (determined by subjective report) the relative weighted RMS values exceeded 18 percent for stimuli NP and DF and were less than 12 percent for stimulus SS. All stereoblind subjects had relative weighted RMS values below 9 percent for all three stimuli. From these data, the criterion value for a VEP to be considered significantly different from a noise signal was set at 12 percent (*13*). There was consistent agreement in the control subjects between the relative weighted RMS values measured and verbal descriptions of the corresponding percepts. The VEP's were therefore assumed to reflect the conscious perception of the stimuli.

Eighteen infants between 7 and 48 weeks of age were selected for study, but one ceased to cooperate after the first

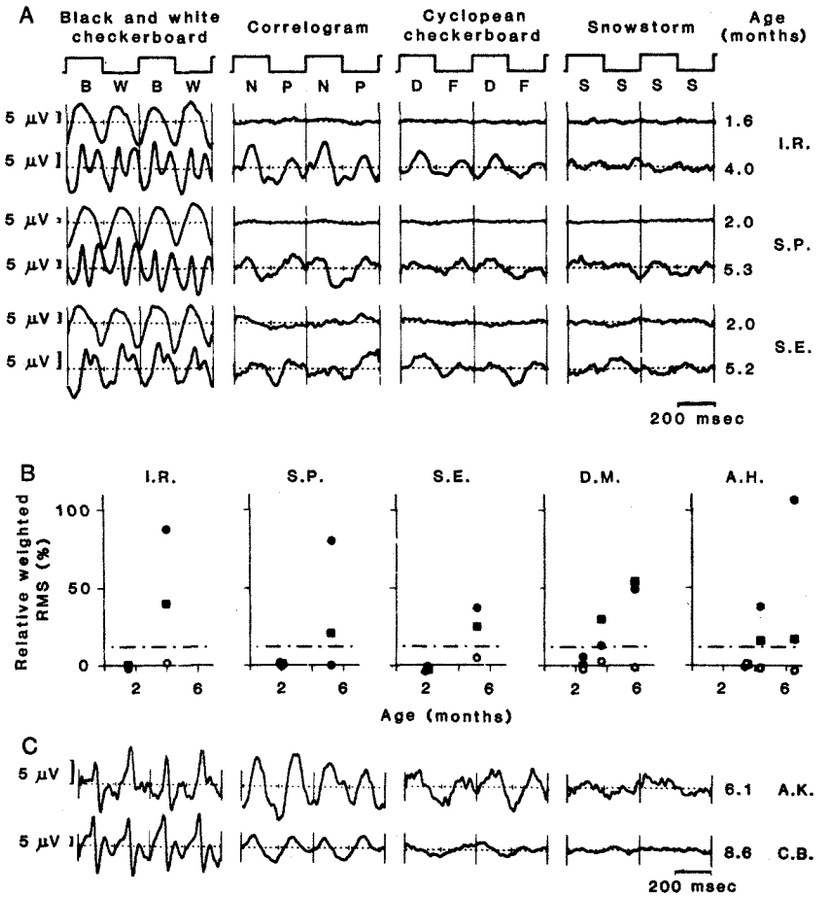

Fig. 2. (A) The VEP's to the four stimuli recorded from three infants (I.R., S.P., and S.E.) at different ages. Dashed line represents computed zero after elimination of the direct-current voltage offset. Positivity at the occipital electrode results in a downward deflection of the trace. The VEP's for each subject are scaled so that the RMS value in the first column (stimulus BW) is unity. Each VEP shows two stimulus periods (delimited by the long vertical lines) each of which consists of two stimulus halves (marked by a short vertical line). (B) Relative weighted RMS values derived from the VEP's of five initially stereoblind infants retested at different ages (see Fig. 1). (C) Same as (A), but for subjects A.K. and C.B. Note doubling of VEP period to NP as opposed to DF.

VEP was measured. The relative weighted RMS values obtained from the mean VEP's of the remaining 17 subjects (Fig. 1B) show a trend from noncriterion responses (< 12 percent) in the younger infants to criterion responses in the older infants. None of the three infants younger than 10 weeks produced criterion responses for the stereoscopic test stimuli, even though they showed acceptable VEP's for control stimulus BW. All eight infants older than 19 weeks in our sample had criterion responses for both the NP and DF test stimuli. Relative weighted RMS values above the 12 percent criterion for both NP and DF were obtained from three of six infants between 10 and 19 weeks. All infants tested had values below the 12 percent for control stimulus SS. The relative weighted RMS values of the youngest infants for test stimuli NP and DF were comparable to those measured in stereoblind subjects (Fig. 1A), but those of the older infants approached the values of stereonormal children and adults (Fig. 1, C and D).

Five of the six infants initially classified as stereoblind were retested later to determine whether the subjects were actually stereoblind, or whether they had been tested before they developed stereopsis. The VEP's of the three youngest subjects (I.R., S.P., and S.E.) (Fig. 2A) show that in their first session the infants produced no VEP's for either NP or DF, but in the second session VEP's were clearly present. Note the development in the VEP for stimulus BW as well. The two older subjects, D.M. and A.H., showed the development of criterion responses also in the second session, and this was confirmed in a third session 2 months later (Fig. 2B).

Some possible causes of our failure to detect criterion VEP's (relative weighted RMS $\geq$ 12 percent) in the very young infants are poor attention; incorrect binocular convergence; image defocus on the retina due to refractive errors, astigmatism, or inadequate accommodation; insufficient visual acuity; and incomplete development of binocular neural functions in the visual system. All but the last probably play minor roles for the following reasons. (i) Looking away from the stimuli was difficult for the infants because of the large back-projection screen we used (*6*). Furthermore, the attention of the infants could be maintained for a sufficiently long time by superimposing a cartoon sound film onto the stimuli in a small region (7° by 5.5° of arc) in the center of the screen. (ii) Correct convergence within Panum's fusional area is necessary for stereopsis (*14*). There is evidence that newborns are able to converge their eyes to binocularly fixate a visual target (*15*), and our target was not physically moving in depth, making it easier to fixate. The cartoon movie in the center of the stimulus also facilitated proper binocular convergence. Further, since the VEP is an average of many measurements, occasional misalignment of the eyes would not make it disappear but only reduce its amplitude. (iii) Although infants 1 to 2 months old may show considerable refractive errors (*16*), the resulting loss of image quality should not prevent the recording of criterion VEP's, since random-dot correlograms and stereograms are known to be particularly insensitive to blurring (*5*). Indeed, we found that criterion VEP's were elicited in an adult control subject after defocusing our stereograms as much as 3 diopters (*17*). (iv) The visual acuity of 1- to 2-month-old infants should suffice to resolve the dots of our patterns (*18*), but, as noted above, this is not essential for perceiving disparate areas in depth or correlation changes.

The most plausible reason for our results appears to be that the neural mechanisms underlying cortical binocularity or stereopsis are not fully developed in the human newborn but start functioning between 10 and 19 weeks. This suggestion is supported by histological findings that the macular region of the retina is immature in newborns (*19*) and that the neural pathways to the visual cortex (*20*) and the synaptic connections in the cortex (*21*) undergo substantial change in the first few months after birth.

We should stress that the stimuli we used cannot efficiently separate cortical binocularity from stereopsis (*22*). However, it was reported (*3*) that in adults VEP's for random-dot correlograms have a double period when compared to those for random-dot stereograms. We observed such doubling in infants beginning at 6 months of age (Fig. 2C).

These findings suggest that the critical period for the development of cortical binocularity or of stereopsis may begin considerably earlier than suggested by binocular tilt transfer studies (*23*). The results from our sample of infants indicate that most infants who are developing normal binocular vision show stereoscopically elicited VEP's when they reach the age of approximately 4.5 months. Our technique allows rapid and objective testing of binocular functions in subjects of all ages. A shortened version of our experimental procedure permits the testing of stereopsis or of cortical binocularity in infants in only a few minutes, and it may provide a useful clinical tool for the early detection and treatment of binocular visual defects and for monitoring the critical period during which cortical binocularity develops.

BENNO PETRIG*
Institute of Biomedical Engineering, University of Zürich, and *Federal Institute of Technology (ETH) Zürich, CH-8044 Zürich, Switzerland*
BELA JULESZ, WALTER KROPFL
Bell Laboratories, Murray Hill, New Jersey 07974
GUENTER BAUMGARTNER
Department of Neurology, University Hospital of Zürich, CH-8091 Zürich, Switzerland
MAX ANLIKER
Institute of Biomedical Engineering, University of Zürich, and *Federal Institute of Technology Zürich*

References and Notes

1. J. Atkinson and O. Braddick, *Perception* **5**, 29 (1976); R. Fox, R. N. Aslin, S. L. Shea, S. T. Dumais, *Science* **207**, 323 (1980); S. L. Shea, R. Fox, R. N. Aslin, S. T. Dumais, *Invest. Ophthalmol. Visual Sci.* **19**, 1400 (1980).
2. D. Lehmann and B. Julesz, *Vision Res.* **18**, 1265 (1978); D. Lehmann, W. Skrandies, C. Lindenmaier, *Neurosci. Lett.* **10**, 129 (1978).
3. B. Julesz, W. Kropfl, B. Petrig, *Proc. Natl. Acad. Sci. U.S.A.* **77**, 2348 (1980).
4. O. Braddick, J. Atkinson, B. Julesz, W. Kropfl, I. Bodis-Wollner, E. Raab, *Nature (London)* **288**, 363 (1980).
5. B. Julesz, *Bell Syst. Tech. J.* **39**, 1125 (1960); *Foundations of Cyclopean Perception* (Univ. of Chicago Press, Chicago, 1971).
6. The dynamic random-dot stimuli were generated by a video random-dot display processor under the control of a PDP-11/40 minicomputer. The stimuli for the left and right eye were separately fed into the green and red channel of a color television projection system (Advent Videobeam 1000A/RGB). Stimuli were back-projected onto a large screen (82° by 68° at a viewing distance of 100 cm) and contained 260 lines of 300 dots each. One dot subtended 20 by 18 minutes of arc. The probability of any dot being either dark (1.5 cd/m^2) or light (15 cd/m^2) was 50 percent at any given time (or television frame). The stimuli were viewed through Kodak Wratten color filters (Nos. 55 and 26) placed both in front of the eyes and the projector guns to achieve a high degree of stereo separation. The binocularly fused green and red images appeared yellowish-white.
7. Dynamic refers to the fact that the entire dot pattern was randomly replaced 50 times per second (television frame rate).
8. D. Regan and H. Spekreijse, *Nature (London)* **225**, 92 (1970); D. Regan and K. I. Beverly, *ibid.* **246**, 504 (1973).
9. Three gold-plated Grass electroencephalogram electrodes were attached to the scalp on the midline with electrode cream: electrode one at 2 cm above the inion, electrode two at 30 percent nasion-inion distance, and a ground electrode at the vertex. The potential difference $V_2 - V_1$ was amplified (gain = 10,000) and band-pass-filtered (−3 dB cutoff frequencies at 0.5 and 40 Hz).
10. The VEP segment was sampled and digitized on the PDP 11/40 minicomputer at a rate of 640 Hz, with 12-bit resolution, in a record of 512 sampled values.
11. The weighting procedure is based on the idea that the VEP (duration T = 800 msec with two full stimulus periods) should ideally be periodic in $T/2$. We defined a weighted RMS value (WR) of VEP(t) by WR = s ρ(0) RMS, where

$$\mathrm{RMS} = \left(\frac{1}{T}\int_0^T \mathrm{VEP}(t)^2\,\mathrm{d}t\right)^{1/2}$$

The zero-lag cross-correlation coefficient ρ(0), which is sensitive to form differences between the two halves of the VEP, is derived from

$$\rho(\tau) = \frac{\frac{2}{T}\int_0^{T/2} \mathrm{VEP}(t)\,\mathrm{VEP}(t + T/2 + \tau)\mathrm{d}t}{(R_1(0)\,R_2(0))^{1/2}}$$

where τ is lag, $R_1(0) = 2/T \int_0^{T/2} \mathrm{VEP}(t)^2\,dt$, and $R_2(0) = 2/T \int_{T/2}^{T} \mathrm{VEP}(t)^2\,dt$. For an ideally repeatable VEP, $\rho(0)$ will approach 1. For a random signal, $\rho(0)$ will be around 0. The other weighting factor is defined as $s = s_e/(s_e + s_o)$ where s_e and s_o are the sum of the even and odd power spectral components of VEP(t), respectively. Calculating the discrete power spectral components of a periodic signal (which has in fact a period of $T/2$) over an integration interval T has the effect that all components at odd multiples of the basic harmonic frequency ($\omega = 2\pi/T$) become zero. For an ideal VEP (no noise, $s_e \neq 0$, and $s_o = 0$), s will approach 1. For a noise signal ($s_e \sim s_o \neq 0$), s will be close to 0.5. Thus, the two weighting factors leave the RMS value of an ideal VEP unchanged, but reduce the contribution of noise to the RMS value of an experimental VEP.

12. The relative weighted RMS values (RWR's) for the three stimuli NP, DF, and SS are defined as
$$\mathrm{RWR(NP)} = 100 \times \mathrm{WR(NP)/WR(BW)}$$
$$\mathrm{RWR(DF)} = 100 \times \mathrm{WR(DF)/WR(BW)}$$
$$\mathrm{RWR(SS)} = 100 \times \mathrm{WR(SS)/WR(BW)}$$
13. Chi-square tests showed no significant differences between the three observed distributions of RWR values (for the stimuli NP, DF, and SS) and three normal distributions with the same mean values and standard deviations. Assuming normal distributions, the probability of a stereoblind subject to produce a RWR value equal to or greater than 12 percent in one of the test stimuli (NP or DF) was estimated as P[RWR(NP) $\geq$ 12 percent or RWR(DF) $\geq$ 12 percent] = .0025. The probability of a stereonormal subject producing a RWR value less than 12 percent in both test stimuli was estimated as P[RWR(NP) < 12 percent and RWR(DF) < 12 percent] = .0018.
14. D. Fender and B. Julesz, *J. Opt. Soc. Am.* **57**, 819 (1967).
15. A. M. Slater and J. M. Findlay, *J. Exp. Child Psychol.* **20**, 248 (1975).
16. O. Braddick, J. Atkinson, J. French, H. C. Howland, *Vision Res.* **19**, 1319 (1979); H. C. Howland, J. Atkinson, O. Braddick, J. French, *Science* **202**, 331 (1978); J. Gwiazda, S. Brill, I. Mohindra, R. Held, *Vision Res.* **18**, 1557 (1978); I. Mohindra, R. Held, J. Gwiazda, S. Brill, *Science* **202**, 329 (1978).
17. See also G. Westheimer and S. P. McKee, *J. Opt. Soc. Am.* **70** (No. 7), 772 (1980).
18. V. Dobson and D. Y. Teller, *Vision Res.* **18**, 1469 (1978); J. Atkinson, O. Braddick, J. French, *Invest. Ophthalmol. Visual Sci.* **18**, 210 (1979).
19. I. Mann, *The Development of the Human Eye* (British Medical Association, London, 1964).
20. P. I. Yakovlev and A. R. Lecours, in *Regional Development of the Brain in Early Life*, A. Minkowsky, Ed. (Blackwell, Oxford, 1967), pp. 3–70; K. Nakayama, *Jpn. J. Ophthalmol.* **11**, 132 (1968); T. L. Hickey, *Science* **198**, 836 (1977).
21. J. L. Conel, *The Postnatal Development of the Human Cerebral Cortex* (Harvard Univ. Press, Cambridge, Mass., 1939–1951), vols. 1–4. For a review of the development of binocularity in cats and monkeys, see J. A. Movshon and R. C. Van Sluyters [*Annu. Rev. Psychol.* **32**, 477 (1981)].
22. Although it was found in adults (*3*) that cyclopean checkerboards with alternating crossed and uncrossed binocular disparities elicit VEP's, they are smaller in amplitude than the VEP's elicited by the cyclopean checkerboards used in our study. For our stimuli, not only the disparity but also the amount of binocular correlation changes. Therefore, the question of whether the cyclopean checkerboard VEP's that we measured are attributable either to cortical binocularity or to the somewhat more complex process of stereopsis cannot be answered [this ambiguity of interpretation applies to all published work on infants as of now (*1, 4*)]. Since we have now explored the range of infants' binocularity, it will be profitable to use alternating crossed-uncrossed binocular disparity stimuli, which have constant correlation and, therefore, have the potential to reveal the onset of stereopsis by itself.
23. M. S. Banks, R. N. Aslin, R. D. Letson, *Science* **190**, 675 (1975); A. Hohmann and O. D. Creutzfeldt, *Nature (London)* **254**, 613 (1975).
24. We thank Y. Lehareinger, V. Corti, and P. Lüthy for technical assistance and D. Lehmann and T. Darcey for their comments on the manuscript.
* Present address: Scheie Eye Institute, University of Pennsylvania, 51 North 39 Street, Philadelphia 19104.

3 November 1980; revised 15 April 1981

Mating Preferences Are Not Predictive of the Direction of Evolution in Experimental Populations of *Drosophila*

Abstract. *The general applicability of two models used in predicting evolutionary directions from asymmetry in reproductive isolation was tested in the laboratory. In mate preference tests with strains of* Drosophila melanogaster *whose ancestral and derived relationships were known, no correspondence was found between sexual isolation and direction of evolution.*

Several different models have been proposed for the origin of premating reproductive isolation during speciation (*1, 2*). Examination of pre- and postmating isolation among a variety of closely related species and incipient species of *Drosophila* indicates that reproductive isolation between any two species or two populations of a species tends to be asymmetrical; that is, to favor one of the two species or populations. This asymmetry provides the basis for two opposing models for predicting the direction of evolution among related species of organisms. Kaneshiro (*3*) proposed that females from an ancestral population discriminate against males of the derived population because derived males have lost important courtship elements and that this is general enough to be considered a rule. Watanabe and Kawanishi (*4*) claimed the opposite, namely, that derived females do not mate with ancestral males and that courtship elements are gained, not lost, during evolution. The authors of the two models assume they know the correct phylogenetic relationships among the species they studied. However, since present-day investigators were not witness to the speciation events, we cannot be absolutely certain which species are ancestral and which are derived. My results in mating tests with populations whose ancestral or derived status with respect to each other is known show that neither the Kaneshiro model nor the Watanabe-Kawanishi model is invariably correct.

Since 1971, a heterogeneous base population of approximately 3000 *Drosophila melanogaster* has been maintained in a population cage at Arizona State University (*5*). From this base population, four derived strains of flies were selected for photopositive, photonegative, geopositive, and geonegative behavior in Hirsh-Hadler mazes (*5*). Three different experiments were carried out in June and July 1980 to determine whether mating preferences between the base and derived strains were consistent with either model. (i) In a series of multiple choice experiments, equal numbers of males and females from the ancestral strain and one derived strain (five pairs from each strain) were placed in an observation chamber for 1 hour. The joint isolation index (*6*) was calculated from the resulting data. (ii) In female choice tests, one female, either ancestral or derived, and two males, one from the same strain as the female and one from another strain, were observed for 1 hour. The female isolation index (*6*) is a measure of the proportion of matings with a male from the same strain as the female. (iii) In male choice experiments, one male and two females, one from the same strain as the male, were observed, and the male isolation index was calculated (*6*). The male and female isolation indices should allow partitioning the joint index into isolation arising from male and female preferences. Isolation indices are significant at $P = .05$ when Z values exceed 1.96 (*4*).

Multiple choice experiments resulted in two significant indices, one of which (that for the photonegative and base strains) reflected positive assortative mating (Table 1). A significant tendency toward heterogametic matings was shown by the photopositive and base strains. Thus the multiple choice experiments have not shown any mating preference patterns that might influence the direction of evolution.

Since females are assumed to choose

Table 1. For each multiple choice test, five pairs of flies from the ancestral (A) strain and five pairs of flies from a derived (D) strain were observed for 1 hour.

Derived strain	Matings observed in multiple choice tests					
	Repetitions	A♀ × A♂	A♀ × D♂	D♀ × A♂	D♀ × D♂	$I \pm$ S.E.
Geopositive	8	20	18	16	24	0.128 ± 0.105
Geonegative	8	18	20	16	18	0
Photopositive	7	17	11	13	28	−0.282 ± 0.096*
Photonegative	10	18	26	24	10	0.304 ± 0.100*

*$P < .05$.

0036-8075/81/0918-1405$01.00/0