tion] and a nonhemolyzing amount of mouse complement (C′). This complex (EAC′) causes rosette formation (*13*) not only with lymphocytes carrying surface membrane immunoglobulin (bursa equivalent or B cells), but with monocytes [by virtue of their surface receptor sites for IgG and complement (*14*)] and with basophils, which carry surface receptors for complement (*15*). The EAC′ rosettes consistently failed to grow on culture, that is, the growth chambers contained only lymphocytes, macrophages, and basophils when examined at periods of up to 4 weeks. Cells that failed to form EAC′ rosettes constituted a single population on velocity sedimentation analysis (Fig. 1B); this population consisted of more than 95 percent lymphocytes, and demonstrated stem cell "activity" on culture.

Mononuclear cells were tested for rosette formation, first with N-SRBC and then with EAC′. Neither rosette product grew in the culture system, but the residual cells, which had failed to form rosettes by either technique, regularly exhibited true pluripotentiality. We believe that this study is the first unequivocal identification of the human hemopoietic stem cell.

RONALD D. BARR
JACQUELINE WHANG-PENG
Division of Cancer Treatment, National Cancer Institute, National Institutes of Health, Bethesda, Maryland 20014
SEYMOUR PERRY
National Institutes of Health, Bethesda, Maryland

References and Notes

1. J. M. Yoffey, *Ciba Found. Symp.* **13**, 4 (1973).
2. J. E. Till, E. A. McCulloch, *Radiat. Res.* **14**, 213 (1961).
3. F. Bohne, R. J. Haas, T. M. Fliedner, I. Fache, *Br. J. Haematol.* **19**, 533 (1970).
4. P. A. Chervenick and D. R. Boggs, *Blood J. Hematol.* **37**, 131 (1971); J. E. Kurnick and W. A. Robinson, *ibid.*, p. 136; K. B. McCredie, E. M. Hersh, E. J. Freireich, *Science* **171**, 293 (1971).
5. D. Zucker-Franklin, G. Grusky, P. L'Esperance, *Proc. Natl. Acad. Sci. U.S.A.* **71**, 2711 (1974).
6. N. Gengozian, *Ann. N.Y. Acad. Sci.* **120**, 91 (1964); W. R. Boecker, A. Boyum, A. L. Carsten, E. P. Cronkite, *Blood J. Hematol.* **38**, 819 (1971); A. Boyum, W. R. Boecker, A. L. Carsten, E. P. Cronkite, *ibid.* **40**, 163 (1972); D. J. P. Squires, *Br. J. Haematol.* **29**, 89 (1975).
7. D. Hoelzer, E. Kurrle, U. Ertl, A. Milewski, *Eur. J. Cancer* **10**, 579 (1974).
8. D. J. P. Squires, L. F. Lamerton, *Br. J. Haematol.* **29**, 31 (1975).
9. A. Boyum, *Scand. J. Clin. Lab. Invest. Suppl.* **21**, 97 (1968).
10. H. P. Lohrmann, L. Novikovs, R. G. Graw, *J. Exp. Med.* **139**, 1553 (1974).
11. L. H. Brubaker and W. H. Evans, *J. Lab. Clin. Med.* **73**, 1036 (1969).
12. R. G. Miller and R. A. Phillips, *J. Cell. Physiol.* **73**, 191 (1969).
13. W. H. Lay, N. F. Mendes, C. Bianco, V. Nussenzweig, *Nature (Lond.)* **230**, 531 (1971).
14. A. F. Lobuglio, R. S. Cotran, J. H. Jandl, *Science* **158**, 1582 (1967); H. Huber, M. J. Pollery, W. D. Linscott, H. H. Fudenberg, H. J. Mueller-Eberhard, *ibid.* **162**, 1281 (1968).
15. M. B. Pepys and A. E. Butterworth, *Clin. Exp. Immunol.* **18**, 273 (1974).
16. We thank E. Lee, A. Brooks, and T. Knutsen for technical assistance.

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Ultrasensitive Chemosensory Responses by a Protozoan to Epinephrine and Other Neurochemicals

Abstract. *A behavioral assay was developed based on differential tendency of a protozoan to attach to an agar gel containing the test substance. The heterotrophic marine dinoflagellate* Crypthecodinium (Gyrodinium) cohnii *responded negatively (less tendency to attach) to epinephrine at concentrations above* 5×10^{-15}M *and to norepinephrine at concentrations above* 5×10^{-9}M. *Response to choline as choline* H_2 *citrate, choline bitartrate, and choline chloride was negative above* 10^{-7}M, *but response to the choline analog carbachol was positive (greater tendency to attach) in the range* 5×10^{-6} *to* 5×10^{-4}M. *Other responses to neurochemicals at comparable concentrations were: dopa, betaine, and glycine—positive;* L-*glutamic acid, tryptophan, putrescine, and taurine—negative. Serotonin was inert, responses to tyrosine and γ-aminobutyric acid were variable, and phenylalanine (*6×10^{-3}M*) and 5-hydroxytryptophan (*5×10^{-4}M*) were negative only at the highest concentrations tested.*

Recent work indicates the importance of chemosensory responses in the ecology of aquatic microorganisms (*1*). We have developed a method for detecting a settling-and-attaching response by the heterotrophic marine dinoflagellate *Crypthecodinium (Gyrodinium) cohnii* to chemicals impregnating an agar gel (*2*). This free-swimming saprophytic species often appears in samples of rotting *Fucus* or other algae (*3*); it grows well on agar slants (*4*) and tends to attach to an agar surface in biphasic culture. This may denote an adaptation for finding and colonizing favorable substrates in nature, and so differential responses to chemicals incorporated in the gel may have an ecological interpretation. Elsewhere (*2*) we report sensitivities to chemicals likely to be present in the vicinity of rotting seaweeds. In an effort to learn the mechanism, or mechanisms, of these responses we have investigated the effects of chemicals that seem unlikely to occur naturally as ecological cues but which are known to be involved in the chemosensitivity of metazoan cells. Here we report extremely sensitive behavioral responses to epinephrine and other neurochemicals and analogs.

Table 1. Concentrations of neurochemicals at which responses occurred.

Chemical	Active molar range
Positive response (greater tendency to embed in experimental gel)	
Dopa	5×10^{-7} to 5×10^{-4}
Betaine	8.5×10^{-6} to 8.5×10^{-5}
Glycine	10^{-6} to 10^{-5}
Carbachol	5×10^{-7} to 5×10^{-4}
Negative response (less tendency to embed in experimental gel)	
Epinephrine	5×10^{-14} to 5×10^{-8}
Norepinephrine	5×10^{-9} to 5×10^{-6}
Choline H_2 citrate	3×10^{-7} to 3×10^{-3}
Choline bitartrate	2×10^{-7} to 2×10^{-3}
Choline chloride	7×10^{-7} to 7×10^{-3}
L-Glutamic acid	7×10^{-8} to 7×10^{-4}
Tryptophan	5×10^{-7} to 5×10^{-4}
Putrescine 2HCl	6×10^{-7} to 6×10^{-3}
Taurine	8×10^{-7} to 8×10^{-3}
5-Hydroxytryptophan	5×10^{-4}
Phenylalanine	6×10^{-3}
Inert (no response detected)	
Serotonin	6×10^{-7} to 6×10^{-3}

In the assay a suspension of organisms in salt solution (*5*) was poured as a fluid overlay onto agar gel in a divided petri dish. Control and experimental gels were made with salt solution and the experimental gel had the test substance added; the two gels were in different partitions of the same petri dish. After standing for 3 hours the suspension was poured off and the agar surface was gently rinsed with distilled water and stained with Lugol's iodine. The densities of embedded organisms remaining in control and experimental gels were compared by counting cells in 40 random microscope fields from each gel and by using Wilcoxon's nonparametric test (*6*). We adopted nonparametric methods because it was clear in all our data that the embedded cells have a clumped distribution with variance of the counts an order of magnitude greater than the mean (*7*). This suggests that the cells may be attracted to each other, as well as to the agar. (Some evidence which we have of such pheromonal activity will be presented elsewhere.) Dead or immobile cells do not embed in the agar (*2*).

By this method we found negative response (smaller density of embedded cells in the experimental partition) to epinephrine at concentrations of 5×10^{-14} to $5 \times 10^{-8}M$. At $5 \times 10^{-15}M$ the effect occurred in some but not all experiments, and at lower concentrations it was not detected. Norepinephrine HCl was negative at 5×10^{-9} to $5 \times 10^{-6}M$. These results and those with other neurochemicals are given in Table 1. Serotonin (6×10^{-7} to $6 \times 10^{-3}M$) was inert (no detected response). Tyrosine and γ-aminobutyric acid are clearly active but experiments to date are somewhat variable. The response to tyrosine seems to depend in part on pH—being negative below pH 6.0, positive above 6.3, and somewhat variable in the pH range 6.0 to 6.3. Response to γ-aminobutyric acid appears to be concentration dependent, being positive at low (10^{-6} to $10^{-5}M$) and negative at high (10^{-4} to $10^{-2}M$) concentrations. However, both chemicals gave somewhat variable results from one experiment to the other; this subject requires further study. Phenylalanine ($6 \times 10^{-3}M$) and 5-hydroxytryptophan ($5 \times 10^{-4}M$) gave negative responses only at the highest concentration tested. Response to the choline analog carbachol (carbamyl choline) was positive in the range 5×10^{-6} to $5 \times 10^{-4}M$.

When the epinephrine threshold is compared to Avogadro's number, it appears that this species is responding to concentrations below 10^6 molecules per milliliter (10^3 molecules per cubic millimeter). We were initially skeptical of these results but repeated trials have consistently borne them out. Possibly the swimming cell explores a sufficiently extensive surface of agar to be influenced by very small concentrations. This hypothesis in turn invites further speculation. It seems likely that stimulation by some minimum threshold number of molecules is needed to evoke the behavioral response but at these concentrations, if the molecules are randomly distributed in the gel, the organism is unlikely to encounter more than one at a time. Perhaps there is a functional "memory" that keeps track of stimulation by molecules encountered during a given time. A plausible mechanism would be a graded series of subthreshold increases in a membrane generator potential, as with nerve cells and some known protozoan responses (*8*).

Most pharmacological studies with protozoa have assayed growth or metabolic responses to much higher concentrations (*9*). Some studies of behavioral responses to chemicals may be related to those reported here; in particular, Sleigh noted stimulation of the motion of ciliate protozoa by epinephrine (*10*).

The specificity of responses is seen in the difference in effects of choline and its analog carbachol. Elsewhere (*2*) we reported a positive response to L-fucose, found in seaweeds, but the synthetic D-fucose is inert. Similarly, responses to β-dimethyl-propiothetin and betaine were positive, whereas the analog dimethyl-β-acetothetin was inert.

These observations indicate the presence of relatively specific chemoreceptors in this species, comparable to those in bacteria (*11*) and both cellular (*12*) and true slime molds (*13*). The great sensitivity to certain neurochemicals seems puzzling from an ecological point of view, but it suggests that *C. cohnii* may prove to be a useful pharmacological tool.

DONALD C. R. HAUSER
M. LEVANDOWSKY
JUDITH M. GLASSGOLD
Haskins Laboratories at Pace University, 41 Park Row, New York 10038

References and Notes

1. I. Chet, S. Fogel, R. Mitchell, *J. Bacteriol.* **106**, 863 (1971).
2. D. C. R. Hauser, M. Levandowsky, S. H. Hutner, L. Chunosoff, J. Hollwitz, *Microb. Ecol.* **1**, 246 (1975); D. C. R. Hauser, M. Levandowsky, S. H. Hutner, L. Chunosoff, *J. Protozool.* **20** (Suppl.), 496 (1973). The tropical "PR" strain was studied.
3. P. Javornicky, *Preslia* **34**, 98 (1962); E. G. Pringsheim, *Farblose Algen* (Gustav Fischer Verlag, Stuttgart, 1963).
4. S. E. Keller, S. H. Hutner, D. E. Keller, *J. Protozool.* **15**, 792 (1968).
5. Medium (in grams per 100 ml): NaCl, 3.0; $MgSO_4$ (anhydrous), 0.35; KCl, 0.08; $CaCO_3$, 0.05; KH_2PO_4, 0.01; pH adjusted to 6.0 to 6.2. These are the major salts, other than substrates, in the chemically defined maintenance medium [see (*2*) and (*4*)]. Ionagar No. 2 should be used.
6. S. Siegel, *Nonparametric Statistics: For the Behavioral Sciences* (McGraw-Hill, New York, 1956). All results reported here are statistically significant by this test with the probability of $\leq .01$ that the result occurred by chance. In most cases we observed $P \leq .001$. Since this was found in replicate experiments, the statistical significance is actually higher. Furthermore, since most of the quantitative information in the data is ignored by nonparametric methods, the reported results may be taken as a conservative interpretation of the data; chemosensory results may have occurred that were not detected by this method (a type II error), but the probability of the reverse (a type I error) is negligible.
7. In a random, or Poisson, distribution the expected values of variance and mean of the samples are equal. A number of indices of aggregation or "clumpedness" based partly on this fact are described in E. C. Pielou, *An Introduction to Mathematical Ecology* (Wiley, New York, 1969). For instance, F. N. David and P. G. Moore [*Ann. Bot. Lond.* **18**, 47 (1954)] studied the "index of clumping" I = (variance/mean) − 1. By their test and others, our data all have very clumped distributions, so that standard (parametric) methods are inappropriate.
8. R. Eckert and Y. Naitoh, *J. Gen. Physiol.* **55**, 467 (1970).
9. S. H. Hutner, H. Baker, O. Frank, D. Cox, in *Biology of Tetrahymena*, A. M. Elliott, Ed. (Dowden, Hutchinson & Ross, Stroudsburg, Pa., 1974), p. 411; S. H. Hutner, *J. Protozool.* **11**, 1 (1964); G. H. Ball [*Univ. Calif. Publ. Zool.* **26**, 353 (1925)] studied effects of vertebrate hormones on the division rate of *Paramecium*.
10. M. A. Sleigh, *J. Exp. Biol.* **48**, 111 (1968); see also the review by S. Dryl, in *Behaviour of Microorganisms*, A. Perez-Miravete, Coordinator (Plenum, London, 1973), p. 16.
11. J. Adler, *Science* **166**, 1588 (1969); I. Chet, Y. Henis, R. Mitchell, *J. Bacteriol.* **115**, 1215 (1973).
12. T. M. Konijn, D. S. Barkley, Y. Y. Chang, J. T. Bonner, *Am. Nat.* **102**, 225 (1968).
13. M. J. Carlile, *J. Gen. Microbiol.* **63**, 221 (1970).
14. We thank Drs. S. H. Hutner, C. Bacchi, C. Beam, and M. Himes for advice and J. Hollwitz, J. Klejman, J. Ruocco, D. Joseph, and M. Romain for technical assistance. Supported by a grant from the S. and W. T. Golden Foundation and by NIH grant GRS FR-05596 to Haskins Laboratories.

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Right Hemisphere Lateralization for Emotion in the Human Brain: Interactions with Cognition

Abstract. *Right-handed subjects tend to look to the left when answering affective questions. The relative shift in gaze from right to left is accentuated when the questions also involve spatial manipulation and attenuated when the questions require verbal manipulation. The data support the hypothesis that the right hemisphere has a special role in emotion in the intact brain, and that predictable patterning of hemispheric activity can occur when specific combinations of cognitive and affective processes interact.*

When a person is asked to picture and describe a happy experience he had, a complex pattern of both cognitive and affective processes is evoked. In the past, such intricate behavioral epochs were studied merely descriptively, since sensitive methods to test relevant process-oriented theories were lacking. However, recent research assessing hemispheric asymmetry in the intact brain has provided a new means of revealing some of the component neuropsychological mechanisms underlying complex, multiprocess tasks. This can be accomplished by comparing the relative activation of the electroencephalogram (EEG) between the two hemispheres (*1*), by assessing performance on a variety of behavioral tasks which reflect hemispheric involvement (*2*), or more simply by observing the direction in which a person shifts his gaze while answering a reflective question (for example, looking to the right is indicative of relative left hemispheric involvement) (*3–8*). When these measures are used with right-handed subjects, verbal and sequential processes and behaviors such as writing a letter or reflecting on a verbal question have typically been associated with a predominance of left hemispheric function, while processes found to be more associated with relative right hemispheric activation include (i) spatial tasks such as a block design test or reflecting on a spatial-pictorial question, and (ii) musical tasks such as identifying instances of a particular theme in an unfamiliar musical selection.

Recently, the right hemisphere has been indirectly implicated as having a special role in the regulation of affective tone (*9*, *10*) although few data in the intact human have been collected to date (*11*). Clinically, patients with unilateral left hemispheric lesions typically respond quite severely to their illness, evidencing more instances of "catastrophic reaction" than do com-