a subcutaneous injection of 3 mg/kg every 6 hours, and were then completely withdrawn from morphine for 3 months. These formerly dependent monkeys had never received nalorphine prior to this study. Two nondependent monkeys that had never been exposed to morphine or nalorphine served as controls.

The monkeys were surgically prepared with chronic jugular catheters (*2*) and they were then trained to press a lever for food; every tenth response was reinforced with a pellet of food (a fixed-ratio of 10 schedule of reinforcement). Each monkey worked on this schedule for 1 hour a day. Four daily sessions were conducted in which saline was given intravenously through the catheter during the food period. After these control sessions, a series of nalorphine doses, administered through the catheter, was tested in ascending order in successive daily sessions during the food period.

Changes in responding, when observed after injections of nalorphine, usually persisted for only several minutes. Only after the 6.4-mg dose of nalorphine (per kilogram of body weight) was given were longer lasting changes in responding observed (Fig. 1). Doses of nalorphine from 0.2 mg/kg to 1.6 mg/kg produced no change in the response rate of the two nondependent monkeys compared to response rate after saline injection. Suppression of their responding was first observed at a dosage of 3.2 mg of nalorphine per kilogram. The three monkeys formerly dependent on morphine showed no change in their response rate after doses of nalorphine from 0.1 to 0.4 mg/kg. Unlike the nondependent monkeys, however, the 0.8 mg/kg and 1.6 mg/kg doses of nalorphine suppressed their response rate. A dose of 3.2 mg/kg of nalorphine, which only partially suppressed the response rate of nondependent monkeys, almost completely suppressed responding of the three formerly dependent monkeys. In addition, emesis, excessive salivation, and hyperirritability were observed in the three formerly dependent monkeys after the 3.2- and 6.4-mg injections of nalorphine per kilogram. In contrast, none of these signs were observed in the nondependent monkeys.

Rhesus monkeys formerly dependent on morphine, with no previous history of nalorphine-induced abstinence, show an increased sensitivity to nalorphine when compared with nondependent monkeys with no history of exposure to morphine or nalorphine. After complete withdrawal of morphine from dependent organisms, certain signs of the morphine-abstinence syndrome persist for many months. This phenomenon has been observed in both animals and man in studies of physical dependence and has been called a secondary abstinence syndrome (*3*). Altered sensitivity to the effects of nalorphine and morphine could be considered part of this secondary abstinence syndrome, and may have implications for the treatment of formerly dependent patients.

STEVEN R. GOLDBERG
CHARLES R. SCHUSTER*
Department of Pharmacology, University of Michigan Medical School, Ann Arbor 48104

References and Notes

1. H. F. Fraser and H. Isbell, *J. Pharmacol. Exp. Ther.* **105**, 498 (1952); J. Cochin and C. Kornetsky, *ibid.* **145**, 1 (1964); C. Kornetsky and G. Bain, *Science* **162**, 1011 (1968).
2. C. R. Schuster and J. V. Brady, *Pavlov J. Higher Nerv. Activ.* **14**, 448 (1964).
3. C. K. Himmelsbach, *Arch. Intern. Med.* **69**, 766 (1942); W. R. Martin, A. Wikler, C. G. Eades, F. T. Pescor, *Psychopharmacologia* **4**, 247 (1963).
4. Supported by NIMH grant 5R-10MH-12084; M. H. Seevers, principal investigator.
* Present address: Department of Psychiatry, University of Chicago, Chicago, Illinois 60637.

11 August 1969, revised 2 October 1969

Neuronal Network Triggering a Fixed Action Pattern

Abstract. *Bursts of impulses in groups of brain cells of the nudibranch* Tritonia *trigger prolonged swimming that is identical to the natural escape response. The cells in which the activity occurs form two bilaterally symmetrical groups of at least 30 cells in each pleural ganglion. These neurons are interconnected by pathways that have a low electrical resistance, both within a ganglion and across the brain. Together they form a network that determines whether a swimming escape response will occur or not by filtering out weak neural activity yet responding with a burst of impulses to intensive specific input to either group.*

It is not yet known what kind of central neuronal events initiate stereotyped behavioral acts, such as fixed action patterns, but the results of ethological studies suggest that the first event is a triggerlike process (*1*). A single brief event lasting no more than a second causes a whole behavioral sequence to occur, and this response may greatly outlast the stimulus. Nothing is known about the neurophysiological basis of this triggering process because of the inaccessibility and lack of knowledge concerning the location of the relevant neurons. In vertebrate animals, a number of instances of elicitation of complex motor acts or sensory experience following stimulation of selected hypothalamic or cortical regions are known (*2*), but these are too complex to permit analysis at the cellular level. Interneurons that drive relatively complex, coordinated movements (*3*) are known in invertebrates, but they must be continually active in order to be effective and do not meet the criteria required of decision-makers. In the intact animal the initiation of activity must have been made ahead of these interneurons, since the stimulus to activity may be a single, brief event.

We located a group of neurons in the brain of the nudibranch mollusk *Tritonia gilberti* which seems to meet the criteria. The cells control the initiation of a fixed action pattern (*4*) of moderate complexity—the swimming escape response. This consists of several sequential, coordinated activities: (i) reflex withdrawal of the oral veil, rhinophores, and branchial tufts; (ii) elongation by contraction of circular body wall muscles and expansion of oral veil and tail; (iii) a series of from one to eight cycles of dorsal and ventral flexions of the body wall musculature; and (iv) one to four spasmodic dorsal flexions. The response is elicited naturally by epidermal contact with certain species of starfish, or it can be evoked by certain surface active agents and salts. Activity of brain cells during the execution of these movements was monitored with intracellular glass micropipettes placed in identified cells of the cerebral-pleural-pedal ganglion complex with the use of an intact animal preparation similar to that described earlier (*5*), except that immobilization of the brain has been improved. In the newer procedure the connective tissue surrounding the brain was pinned to a small wax-covered table held on the end of a rigid arm, placed beneath the brain.

The cells whose coordinated activity triggers swimming are located in two groups [numbered 10 and 21 in earlier

publications (*5*)], symmetrically located on the pleural ganglia near the central commissure. They are small compared with others in the ganglion, having diameters of 50 to 150 μ; their peripheral axons pass into the ipsilateral main pleural nerve (PN1) and pedal nerves (PeN1 and PeN2) (*6*). In addition to their trigger action, the neurons have a motor function.

When stimulated singly by means of intracellular electrodes, the neurons cause contractions of body wall musculature that appear to be a part of phases (i) and (ii) of the response. They are excited by fibers in all the major nerve trunks from epidermal receptors distributed over the whole surface of the body. The neurons extend around the medial surfaces of the pleural ganglia close to the central commissure and may continue as far as the ventral surfaces, as well as through the core, so that an accurate upper limit on the total number of cells cannot yet be given. However, there are approximately 30 cells visible in each of the dorsally located groups.

The trigger function of the neurons can be demonstrated in five ways: (i) Occasionally, direct stimulation of a single cell in one of the groups with a current just sufficient to produce a single impulse or short burst leads to a swimming escape response (*5*). (ii) Brief stimulation applied through a suction electrode, large enough to cover many surface cells in either group, invariably elicits the entire behavioral sequence (Fig. 1b). Similarly, gentle tapping of the surface of the brain over the groups evokes the full response. (iii) In isolated brain preparations, stimulation as in (ii) is followed by sequences of patterned impulse activity in other neurons that closely resemble those seen in the same cells during the swimming escape response in an intact preparation (*7*). (iv) When the escape response is initiated through normal sensory modes it is always preceded by at least one burst of impulses in cells of the groups (Fig. 1a). (v) In isolated brain preparations, stimulation of cerebral nerve trunks sufficient to cause patterned activity in neurons associated with swimming always produces a burst in the cells of the groups.

Unique and functionally critical characteristics of the neurons involved in the trigger action are that they are all interconnected and that these connections have a low electrical resistance.

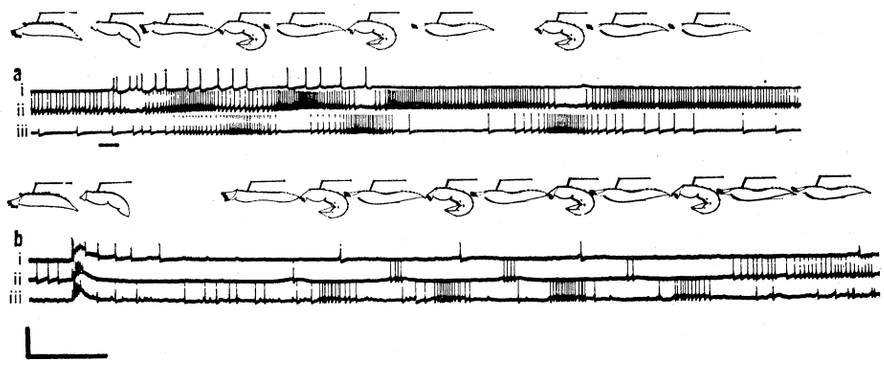

Fig. 1. Initiation and execution of the swimming escape response. Intracellular electrodes were placed in the following neurons: *i*, cells of the decision-making, trigger group (right pleural ganglia); *ii*, cells driving dorsal flexion; and *iii*, cells driving ventral flexion. Swimming escape responses were elicited in (a) by dropping a few crystals of sea salt onto the oral veil (moment of application indicated by bar) and in (b) by applying six impulses at 10 per second to several trigger group cells simultaneously, through a suction electrode placed over them; (a) and (b) are two different preparations. Movements of the animal associated with the neural activity are shown by the small drawings. Further details in text. Calibration: 100 mv, 5 seconds.

When a current pulse, either depolarizing or hyperpolarizing, is passed into one, this spreads not only to its neighbors but also to members of the group in the other pleural ganglion, across the commissure (Fig. 2, a and b). A consequence of the interaction that occurs between the ganglia, in either direction, is that any unilateral inputs contribute to bilateral trigger group activity. This may help to coordinate the observed bilateral response of the whole animal. The extent of the coupling between immediately adjacent cells was found to be extremely high and to fall off with increasing distance from the stimulated neuron. The excitation of any neuron within either group results in graded stimulation of many others. Depolarizing waves and action potentials therefore occur at about the same time in cells of the trigger group.

The interconnectedness is further demonstrated if there is a delay in the firing of one of the somas of a cell of the group (*8*), as occurs in the example shown in Fig. 2b. The late action potential feeds back into neighboring cells and could re-excite them. Thus it might be imagined that almost any activity initiated within the network would start up the bursting and lead to swimming, but this is not the case. The excitability of the network is determined by the summed activity in all of the component neurons. Only when this excitability is high overall can there be a cascading effect leading to the generation of bursts of action potentials associated with swimming. An example

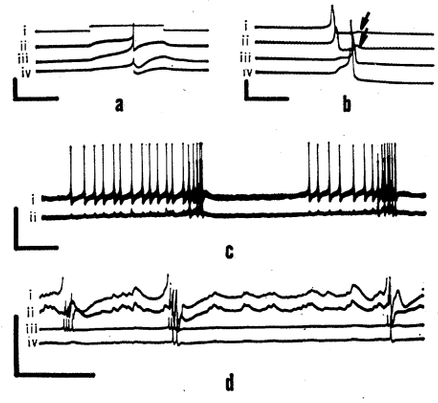

Fig. 2. Characteristics of neurons of the decision-making, trigger group: (a) d-c coupling between neurons. Four intracellular electrodes were placed into different cells of the group—*i*, *ii*, and *iii* in cells of the right pleural ganglion; *iv*, in a cell of the left pleural ganglion. A current pulse was applied to *i* (artifact monitored at lower gain than *ii* to *iv*) and produced depolarization and an impulse not only in *i* but also in *ii*, *iii*, and *iv*. (b) Feedback within the network. Recordings of spontaneous firing in the same four cells as in (a), but at a higher sweep speed. Impulses in *i* and *ii* are nearly simultaneous; soma responses in *iii* and *iv* are delayed, then feed back into *i* and *ii* (arrows). (c) Spontaneous cascading in trigger network. Accelerating bursts resulting from coupling and feedback. (d) Common sensory inputs. The oral veil was touched by a starfish, leading to similarly phased inputs of comparable amplitudes in two different neurons of the group; *i* and *ii* are the same as *iii* and *iv* but at higher gain. Records (a) to (c) are from isolated ganglia, and (d) is from an intact animal preparation. Calibration: vertical, (a) to (c) and *i* and *ii* in (d), 50 mv; *iii* and *iv* in (d), 250 mv; horizontal, (a) and (c), 2.5 seconds; (b), 0.25 second; and (d), 1 second.

of the initiation of a bursting under conditions of high excitability is given in Fig. 2c.

A clue to understanding the mode of operation of the trigger group came as a result of studying the response to natural stimuli that elicit swimming activity. Recordings were made from many neurons in the network, with as many as four electrodes recording simultaneously from different cells. Contact with one or a few tube feet of a starfish was followed by bursts of small excitatory postsynaptic potentials (EPSP's) in all cells recorded from. Surprisingly, some of these were very nearly synchronous in many of the cells (Fig. 2d). The synchrony is not due to electrical conduction since the magnitudes of the EPSP's were similar. It is more likely that inputs from the appropriate receptors arrive at many cells of the network simultaneously owing either to an extensive degree of divergence of sensory neurons before they terminate on the cells of the network or to the interposition of interneurons that provide for the divergence.

These synchronous small depolarizations, as well as weak currents applied by suction electrodes, are effective in giving rise to bursts, provided they occur in a sufficiently large group of neurons at the same time. Large depolarizations of single or small groups of neurons rarely lead to a synchronous burst. The distinction between the effectiveness of synchronous depolarizations in a large number of cells and the ineffectiveness of isolated large depolarizations in leading to bursts may well provide the neuronal mechanism for the trigger action. This type of operation combines aspects of the randomly connected neuron model of Beurle and the "multiple input, metastable feedback loop" of Bullock (*9*).

Occasionally, the animals do execute full swimming sequences in the apparent absence of any external stimulation. These are comparable to the familiar "vacuum activities" observed by ethologists (*1*). In *Tritonia*, they appear to arise as a result of cascading of spontaneous activity by local reexcitation and spreading over the network, at times when there is a high level of "background" activity.

The conclusions from the present evidence are that the function of initiating the swimming escape response resides with the pleural network, and that the basis of its operation is positive selection of inputs that affect many cells in the network simultaneously and rejection of those that excite only a few components.

A. O. D. WILLOWS*
G. HOYLE

Biology Department,
University of Oregon, Eugene 97403

References and Notes

1. N. Tinbergen, *The Study of Instinct* (Oxford Univ. Press, London, 1951).
2. W. Penfield, in *Handbook of Physiology*, sect. 1, *Neurophysiology*, J. Field *et al.*, Eds. (Williams and Wilkins, Baltimore, 1960), vol. 3, pp. 1441–1445; B. Andersson, *Acta Physiol. Scand.* **28**, 188 (1953); F. Strumwasser and T. J. Cade, *Anat. Rec.* **128**, 630 (1957); E. von Holst and U. Von St. Paul, *Naturwissenschaften* **47**, 409 (1960); W. R. Hess, *Hypothalamus and Thalamus* (Thieme, Stuttgart, 1956).
3. K. Ikeda and C. A. G. Wiersma, *Comp. Biochem. Physiol.* **12**, 107 (1964); D. Kennedy, W. H. Evoy, J. T. Hannawalt, *Science* **154**, 917 (1966).
4. W. H. Thorpe, *Learning and Instinct in Animals* (Harvard Univ. Press, Cambridge, 1963).
5. A. O. D. Willows, *Science* **157**, 570 (1967); ———, in *Physiological and Biochemical Aspects of Nervous Integration*, F. Carlson, Ed. (Prentice-Hall, Englewood Cliffs, N.J., 1967), pp. 217–243; ——— and G. Hoyle, *Symp. Biol. Hung.* **8**, 443 (1968).
6. A. O. D. Willows, thesis, University of Oregon (1967), pp. 54–56.
7. D. A. Dorsett, A. O. D. Willows, G. Hoyle, *Nature*, in press.
8. L. Tauc, *J. Gen. Physiol.* **45**, 1077 (1962).
9. R. L. Beurle, *Phil. Trans. Roy. Soc. London Ser. B Biol. Sci.* **260**, 55 (1956); T. H. Bullock and G. A. Horridge, *Structure and Function in the Nervous Systems of Invertebrates* (Freeman, San Francisco, 1965), pp. 283–284.
10. Supported by NSF research grant GB 7413 to G.H. Special thanks are extended to Lee H. Vernon for technical assistance, and to Dr. Robert L. Fernald, director of the Friday Harbor Laboratories, for generous facilities placed at our disposal.

* Present address: Department of Zoology, University of Washington, Seattle 98105.

27 October 1969

Caribbean Cores P6304-8 and P6304-9: New Analysis of Absolute Chronology. A Reply

We are considerably annoyed with *Science* for having published a comment (*1*) critical of our work without informing us of its acceptance and giving us the customary option to reply in the same issue. In this comment, Broecker and Ku report their Th^{230}/Pa^{231} results on the same or adjacent samples from the Miami cores P6304-8 and P6304-9 which we had previously analyzed (*2*). Of the four samples tested, agreement between Miami and Lamont-Doherty was found for two, disagreement for the other two (Table 1 in *1*). Where disagreement exists, Broecker and Ku claim that their results are right and ours are wrong. We believe exactly the opposite and for very good reasons: (i) Our results match exactly the C^{14} time scale over the common range, while theirs do not; and (ii) our results are internally consistent in every case while theirs are not. Thus, not only do Broecker and Ku obtain ages approximately 30,000 years too old for the top of the cores, which are demonstrably modern by both C^{14} (*2*) and micropaleontological analysis at the subspecific level (*3*), but they also find a Th^{230}/Pa^{231} value of 29 for one core level and a value of 40 for a stratigraphically identical level in the other core, while we obtained values of 29 for both cores (Table 1 in *1*).

Our extraction technique (*4*) is directed toward removing from the sedi-

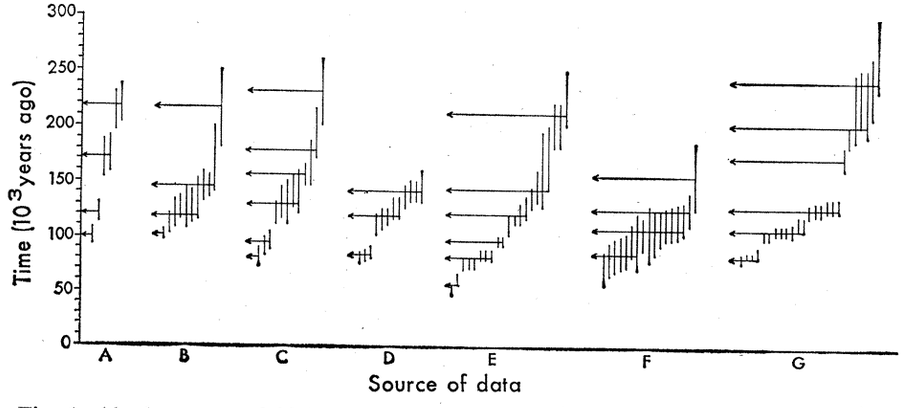

Fig. 1. Absolute ages of high sea levels during the past 300,000 years. Vertical lines are absolute ages with their published errors (from *9–15*) (see Table 1). Horizontal arrows represent best fits (chronological means).