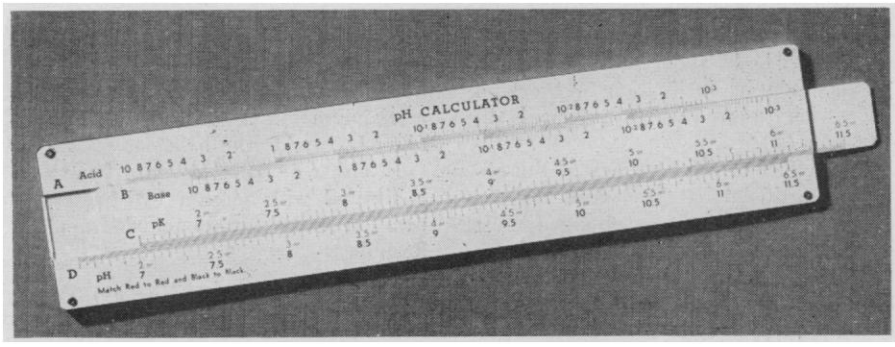

Fig. 2. A pH calculator constructed from linear transformations of the Henderson-Hasselbalch equation. The ratio B/A at this position is 8/3 or 2.67. For any univalent acid the value of $(p\mathrm{H} - pK)$ is then 0.43.

mic paper. The use of two-cycle paper permits the curve to be plotted over a range of two pH units, with the midpoint at pK (Fig. 1). In this diagram $(pA - pB)$ is represented on a linear scale parallel to the logarithmic scale of B/A.

By a further transformation, a semilogarithmic slide rule has been constructed (Fig. 2) (*1*). In this construction, A and B are identical logarithmic scales of acid and base concentrations. These scales are accurately calibrated in four logarithmic cycles at concentrations from $10^{-3}M$ to $10M$. These limits represent values of pA and pB of 3 and -1, respectively. Scales C and D are identical linear scales of pK and pH, respectively. A distance on these scales of 1 pH or pK unit is equal to 5 cm. The scales are calibrated to 0.01 pH, corresponding to 0.5 mm per scale division. The length of one logarithmic cycle on scales A and B is likewise 5 cm; this spacing permits concentrations to be read to two or more significant figures. On the slide rule, scales A and D are fixed, while B and C are movable. The standard position, where acid and base concentrations are equal, corresponds to the midpoint of any titration curve. In this position pH and pK are also equal.

When scales B and C are moved toward the right, the value of $(pA - pB)$ in any position becomes positive and equal to $(p\mathrm{H} - pK)$. Positive values are obtained for the upper half of the titration curve, where B is greater than A, negative values for the lower half of the curve. In all displacements, positive or negative, $(pA - pB)$ is equal to and of the same sign as $(p\mathrm{H} - pK)$. At all positions of scales B and C on scales A and D, the Henderson-Hasselbalch equation is satisfied. These properties are evident from the nature of the linear transformation (Fig. 1).

By the use of an accurate slide rule, any point of a titration curve may be determined in a few seconds. If pK of the buffer is known, the pH of a solution is determined by placing the concentration of base under the concentration of acid. The eight or ten points required for an accurate sigmoid curve may be computed within a minute or two. The converse problem is that of finding the concentrations of acid and base required to yield a buffer of given pH. In this case the value of pK is placed over pH; base concentration is read under acid concentration. In the illustration (Fig. 2), pK of acetic acid, 4.7, is placed on the desired pH, 5.13. At any part of the concentration scales the ratio B/A is 8/3, or 2.67. This number is approximately the antilog of 0.43, the difference between pH and pK. With the slide rule, tables of logarithms or graphs are not required, and calculations of this kind may be made in a few seconds.

NORMAN R. JOSEPH
Department of Chemistry, College of Pharmacy, University of Illinois, Chicago

Reference

1. N. R. Joseph, *Federation Proc.* 17, 685 (1958).

2 June 1958

Property of Cerebrospinal Fluid Associated with Disturbed Metabolism of Central Nervous System

Abstract. Cerebrospinal fluid was assayed for the capacity to contract smooth muscle and for the capacity to develop such activity when incubated with globulin. Activity was observed in fluid collected from patients with inflammatory or degenerative disease of the central nervous system, sustained intracranial vasodilatation, sustained noxious stimulation, or chronic schizophrenia. Control specimens lacked measurable activity.

Cerebrospinal fluid was collected by lumbar puncture from subjects with (i) active disease of the central nervous system (CNS) (recent cerebrovascular accident, neoplasm of the central nervous system, cerebral atrophy, multiple sclerosis), (ii) inactive nonprogressive or no disease of the central nervous system (myasthenia gravis, progressive muscular dystrophy, "old" cerebrovascular accident), (iii) vascular headache of the migraine type, (iv) sustained noxious stimulation and pain arising from disorders of the legs or pelvic organs, (v) disease syndromes classified as chronic schizophrenia (all were hospitalized males between 20 and 50 years of age; they were free of other significant disease and were not overactive or assaultive; some had experienced hallucinations and most had delusions; all were ambulatory and none exhibited evidence of dietary insufficiency; none had had induced convulsions in the year previous to study). Specimens either were assayed at once or were immediately stored in solid carbon dioxide.

The spinal fluid so collected was assayed for its capacity to contract smooth muscle (isolated rat uterus or guinea pig ileum suspended in a 10-ml saline bath at 29°C). The sensitivity of the preparation was assessed by the response of the muscle to 0.2 ml of a polypeptide (bradykinin) standard prepared by the action of trypsin on globulin. (If the response to this material was inadequate, the preparation was discarded.) Freshly collected or thawed cerebrospinal fluid (0.2 ml) was added to the chamber. If contractions were observed the specimen was recorded as inducing a type I reaction. If no contractions were observed, 0.2 ml of the specimen was incubated with 0.2 ml of an 8 percent solution of bovine globulin (fraction II) for 3 minutes at 29°C. If the resultant mixture induced contractions, the specimen was recorded as inducing a type II reaction. If no contractions were observed, the specimen was recorded as negative. The results of this bioassay are shown in Table 1.

The contractions so induced by cerebrospinal fluid or by the incubated mixtures were not inhibited by atropine, antihistaminics (histamine contracts guinea pig ileum but not rat uterus), or dihydroergotamine in amounts large enough to inhibit contractions induced by acetylcholine, histamine, or serotonin, respectively. They were inhibited by slightly larger amounts of dihydroergotamine, by lysergic acid diethylamide, by salicylate, and by soluble adrenal steroids. Prolonged (1 hour) incubation with plasma diminished the response.

Cerebrospinal fluid or incubated mixtures that induced contractions of the smooth muscle also induced pain when applied to an exposed blister base, lowered the blood pressure of a cat when injected into the venous circulation, dilated minute vessels when applied to the bulbar conjunctiva, and increased capillary permeability as indicated by increased spreading of dye in the region surrounding an intradermal injection of the material in a guinea pig with dye injected into the venous circulation.

These properties are similar to those of vasodilator polypeptides derived from

plasma proteins. Keele *et al.* (*1*) demonstrated that such polypeptides were present in blister fluid, in fluid collected from painful joints, and in inflammatory pleural fluid. Ostfeld *et al.* (*2*) demonstrated that a similar substance is present in tissue fluid removed from regions of local tenderness in the scalp during vascular headache of the migraine type. Hilton and Lewis (*3*) described a proteolytic enzyme present in saliva and in saline perfusate of the salivary gland which formed vasodilator polypeptides when incubated with plasma proteins. These authors observed increased amounts of enzyme during heightened metabolic activity of the gland and concluded that the enzyme and polypeptides are responsible for local vasomotor control in the salivary gland. The properties of the polypeptides corresponded to those of "bradykinin," the name given by Rocha é Silva, Beraldo, and Rosenfeld (*4*) to the substance or substances produced on incubation of certain snake venoms or trypsin with serum, plasma, or plasma globulin. These authors reported that when such incubated mixtures were applied to smooth muscle (isolated rat uterus or guinea pig ileum), contractions occurred that were delayed, slow, and sustained.

It therefore was inferred that the specimens which gave type II reactions contained detectable amounts of a proteolytic enzyme capable of acting on plasma proteins (globulin) to form polypeptides of the bradykinin type and that the specimens which gave type I reactions contained detectable amounts not only of the enzyme but of the polypeptides as well.

From these studies it was concluded that increased amounts of "protease" and "vasodilator polypeptides" in the cerebrospinal fluid are associated with: (i) active inflammatory or degenerative disease of the central nervous system, (ii) vascular headache of the migraine type (sustained vasodilation), (iii) prolonged noxious stimulation and pain (sustained excitation within the central nervous system), and (iv) chronic schizophrenic reactions.

Table 1. Results of bioassay.

Condition of patient	Type I	Type II	Negative
Active disease of CNS	8	36	2
Inactive or no disease of CNS	0	2	39
During migraine attack	3	14	1
Seven days after migraine attack	1	0	7
Pelvic or leg surgery (no pain)	0	2	11
Pelvic or leg surgery (sustained pain)	2	9	1
Chronic schizophrenia	4	35	2

A provisional formulation of these findings has been made: During periods of increased or faulty metabolism in the central nervous system, the augmented catabolism results in the accumulation of protease and possibly protein and protein breakdown products in the extracellular and cerebrospinal fluid. Through proteolysis either within or outside minute vessels, this protease forms vasodilator polypeptides that act with other mediators to enhance local blood flow. When appropriate in amount, this response to increased catabolic products serves to meet increased tissue needs by enhancing nutrient supply. When for whatever reason the accumulation of extracellular protease is excessive or there is excessive formation of vasodilator polypeptides, the enhanced capillary permeability and damage to tissue may interfere with the functional capacity of the central nervous system.

It has been shown previously in this laboratory (*5*) that subjects with disease classified as chronic schizophrenia exhibit a fall-off in highest integrative functions in many ways resembling that observed in subjects with serious brain damage. Impairment of highest level brain functions was also observed in subjects with prolonged adaptive difficulties and severe anxiety. The latter subjects, as well as many of those with schizophrenia, were referred to as being in the "unadapted state," a term employed to refer to the totality of reactions (physiological, behavioral, and attitudinal) present in subjects who for long periods have perceived themselves to be severely or overwhelmingly threatened and have been unable to achieve adequate overall adaptation. Prolonged disturbances in the modulation of excitation and inhibition within the central nervous system is inferred to be an aspect of the unadapted state.

It therefore is of special interest that the cerebrospinal fluid of patients with schizophrenia contains an abnormal amount of protease, as also was found in those with active inflammatory and degenerative diseases of the central nervous system and in those in whom a prolonged state of excitation within the central nervous system may be inferred. This observation suggests that a significant (yet perhaps reversible) alteration in metabolism occurs in the brain of patients with schizophrenia. The available evidence does not allow inferences as to whether the accumulation of protease and polypeptides in the cerebrospinal fluid is a manifestation of deranged metabolic function of the brain or is a causative factor in such derangement. However, whether primary or secondary, this accumulation could contribute to impairment of the functions of the brain.

It is suggested that the protease-polypeptide system is implicated in local vasomotor control within the central nervous system and when in excess the components of the system may be relevant to disease.

LORING F. CHAPMAN
HAROLD G. WOLFF
Departments of Medicine and
Psychiatry, New York Hospital–Cornell Medical Center, New York

References and Notes

1. D. Armstrong, C. A. Keele, J. B. Jepson, J. W. Stewart, *J. Physiol. (London)* 135, 350 (1957).
2. A. M. Ostfeld, L. F. Chapman, H. Goodell, H. G. Wolff, *Psychosomat. Med.* 19, 199 (1957).
3. S. M. Hilton and G. P. Lewis, *Brit. Med. Bull.* 13, 189 (1957).
4. M. Rocha é Silva, W. T. Beraldo, G. Rosenfeld, *Am. J. Physiol.* 156, 261 (1949).
5. L. F. Chapman, W. N. Thetford, L. Berlin, T. C. Guthrie, H. G. Wolff, *Proc. Assoc. Research Nervous Mental Disease* 36, 491 (1956).

29 August 1958

Electric Response of Glia Cells in Cat Brain

Abstract. An experiment is described suggesting that the glia cells in the mammalian cerebral cortex are capable of developing electric responses to direct stimuli. When hyperfine microelectrodes were pushed into the cortex of a cat, slow, reversible potential variations were recorded which resembled the "electric responses" from the glia cells in tissue culture.

In recent years, the technique of recording electric responses of single cells with intracellular microelectrodes has been employed by many physiologists working on the mammalian central nervous system. There has been, however, some ambiguity in this type of investigation with respect to the exact position of the recording microelectrode relative to the single unit recorded, or, in many cases, even with respect to the type of cell impaled by the microelectrode. This ambiguity can be completely eliminated if one uses the material in tissue culture and carries out electrophysiological observations under direct visual control.

In November 1957, we were given an opportunity to travel to the Tissue Culture Laboratory of the University of Texas, Galveston, with necessary electric equipment, and to work in collaboration with W. Hild on in vitro nervous elements taken from the cat cerebrum and cerebellum. It was found in this investigation (*1*), in the first place, that both nerve cells and astrocytic glias give sizable resting potentials on penetration with a microelectrode. In response to direct electric stimulation (with an extracellular electrode), nerve cells produced action potentials of duration of