Learning and Behavior (I): Effects of Pituitary Hormones

The nervous and endocrine systems are known to cooperate in regulating the body's activities. For the last few years, the nature of the interactions between them has been a target of intense—and, at times, highly competitive—investigation. One of the products of these studies is an accumulating body of evidence that indicates that a number of peptide hormones act directly on the brain to affect learning or behavior. Consequently, some of the agents are now being tested clinically to see if they are of any value in treating a variety of conditions, including Parkinson's disease, schizophrenia, depression, and learning disorders.

The hormones that have attracted the greatest attention so far are adrenocorticotrophic hormone (ACTH) and melanocyte-stimulating hormone (MSH), both of which are synthesized in the pituitary gland; vasopressin, which is synthesized in a portion of the brain called the hypothalamus but is transported to the pituitary gland where it is stored until needed; and certain factors or hormones that are produced in the hypothalamus. These latter peptides are best known for their roles in controlling hormone secretion by the pituitary. Their behavioral effects will be discussed in a later article.

Much of the evidence concerning the effects of the pituitary hormones on brain function comes from conditioning experiments with animals. The conditioning experiments are of two main types. In avoidance conditioning, the animal must learn to perform a specific action, such as running from one compartment of a shuttle box into the other, in order to escape an unpleasant experience such as an electric shock. In appetitive or approach conditioning, the animal must acquire the behavior in order to achieve a reward such as food, drink, or sex. This type of conditioning is not considered as stressful to the animal as the avoidance type.

In an early experiment, David de Wied and his colleagues at the University of Utrecht in the Netherlands found that removal of the pituitary gland of rats (hypophysectomy) decreased the ability of the animals to acquire conditioned avoidance behavior. Administration of ACTH, MSH, or vasopressin to the hypophysectomized rats, however, corrected this deficiency with the result that these animals learned nearly as well as the controls. In other experiments, the investigators showed that the hormones also delayed the extinction of conditioned avoidance behavior; that is, animals treated with the peptides remembered the task longer than did untreated animals.

(The many investigators collaborating with de Wied on these studies of the effects of pituitary hormones on behavior and learning include Bala Bohus, Willem Gispen, Tjeerd van Wimersma Greidanus, and A. Witter, all at the University of Utrecht, and H. Greven, H. Rigter, and H. van Riezen of the Organon Company in the Netherlands.)

The next step was to determine whether the three hormones acted in the same manner or whether they affected different aspects of the learning process. It was expected that the effects of ACTH and MSH would be very similar because the amino acid sequences of these peptides have a number of similarities. In particular, there is one sequence of seven amino acids that is found in ACTH and in all MSH molecules (there are several, depending on the species). The amino acid sequence of vasopressin, on the other hand, in no way resembles those of ACTH or MSH.

Hormonal Differences

According to de Wied and his colleagues the effects of vasopressin on conditioned avoidance behavior did differ from those of the other two hormones. Injection of ACTH or MSH appeared to delay the loss of avoidance responses by intact rats for a few hours, whereas vasopressin delayed it for several days or longer. This difference could not be explained on the basis of the lifetimes of the hormones because they all have half-lives of only a few minutes. Thus they hypothesized that ACTH and MSH affected motivational processes and that vasopressin affected long-term memory.

Additional evidence supporting the hypothesis that vasopressin is involved in consolidation of long-term memory comes from experiments with rats having a hereditary disease characterized by lack of vasopressin. The Utrecht investigators found that these animals could learn an avoidance behavior but that they forgot it very rapidly unless given vasopressin. Then they remember it as well and for as long as normal controls. Furthermore, injection of antiserum against vasopressin directly into the brains of normal rats, either shortly before or after a training session, prevents the animals from acquiring a long-term memory of the behavior that they learn. Antiserum to oxytocin, a peptide hormone with a structure similar to that of vasopressin, had no effect on long-term memory.

The behavioral effects of the hormones are independent of their better known endocrine effects. ACTH stimulates the production of steroid hormones by the cortex of the adrenal gland. Since these adrenal hormones enable the animal to better cope with stress—"fright, fight, or flight" situations—and generally make the animal more alert and active, it was possible that the effects on learning were the consequence of the activities of the adrenal steroids. However, ACTH and MSH have similar effects on behavior despite the fact that MSH has almost no activity in stimulating the adrenal gland. And de Wied has found that adrenal steroids actually facilitate the extinction of conditioned responses; that is, their effects are the opposite of those of ACTH. Moreover, fragments of the ACTH molecule that have little or no activity in eliciting formation of adrenal corticoids in vivo are as effective in the behavioral assays as the intact molecules. One such fragment, which consists of amino acid residues 4 to 10 of the ACTH molecule and is thus called ACTH 4-10, contains only seven amino acids—the same sequence that is found in MSH molecules.

De Wied says that this sequence contains the information essential for the behavioral effects of these hormones. Changing the sequence even slightly can alter the effects of ACTH 4-10 and even reverse them. For example, replacement of the normal L-isomer of phenylalanine with D-phenylalanine, its mirror image, produces a peptide that facilitates, rather than prevents, loss of conditioned behavior. On the other hand, some changes may potentiate the effects of ACTH 4-10. These include oxidation of methionine to the sulfoxide, and replacement of arginine with D-lysine and of tryptophan with phenylalanine. Combination of all these amino acid substitutions in one molecule produces a peptide that is 1000 times more active than ACTH 4-10. At least part of the enhanced activity may be due to the increased resistance of the altered peptide to degradation by enzymes that break down peptides.

Vasopressin is also released in response to stress. Its major function include maintaining the water balance of the body and increasing the blood pressure. Here again, some analogs of vasopressin that lack these activities are also active as the normal hormone in consolidating long-term memory.

In his early work de Wied focused on the

effects of ACTH and MSH on conditioned avoidance behavior. A group of investigators in New Orleans, including Abba Kastin of the Veterans Administration Hospital there, Curt Sandman, who is now at Ohio State University, and Lois Stratten of the University of New Orleans, began their work on MSH with appetitive tasks in order to avoid the extra stress imposed on animals by aversive conditioning. But they have also used avoidance conditioning, and, in recent experiments, the Utrecht group has studied the effects of pituitary peptides on appetitive behavior. In general, the results of the two groups are in agreement for both types of experiment.

Kastin and Sandman, however, think that MSH and ACTH facilitate learning by improving the animal's ability to pay attention to the task rather than by acting on memory processes. They cite the results of task reversal experiments in support of this position. In task reversal experiments the subject (whether animal or human) must learn to make a response to a particular property—a color, for example. Then the experimenter changes the correct answer. The shift may be to a different color (intradimensional shift) or to a new category (extradimensional shift) such as shape. Kastin and his colleagues trained rats to run to a lighted door in one arm of a Y-shaped maze in order to escape an electric shock. Then they made the dark door the escape route from the shock. Rats treated with MSH learned this reversal faster than those receiving the control solution.

According to Kastin, this result is inconsistent with the theory that MSH increases retention of an acquired behavior because, if that were the case, reversal should be slower, not faster, when the animals are given the hormone. But the result supports the hypothesis that MSH improves the rats' ability to pay attention. A highly attentive rat should learn that the lighted door is the correct escape route and also that the degree of brightness is the key characteristic. This should enable it to make the shift to the dark door more readily than an inattentive rat that had not learned what quality to look for.

Although trials to determine the effects of pituitary peptides in humans are not yet very far along, there is evidence that they do affect learning behavior in humans. Lyle Miller of Temple University, working in collaboration with the New Orleans investigators, found that ACTH 4-10 improved the visual memory, as determined by the Benton visual retention test, and decreased the anxiety of volunteers receiving the drug when compared with those injected with a dilute salt solution. In an earlier study, they obtained similar results with MSH. The peptide did not affect the volunteers' verbal retention nor did it have any effect on their retention times.

When the investigators determined the effects of ACTH 4-10 on the electroencephalograms (EEG's) of the subjects, they observed changes indicating that the peptide prolongs the subject's period of mental alertness. When individuals are exposed to a new stimulus of some kind, the alpha patterns of their EEG's, which are generally associated with a relaxed mental state, are normally replaced with a pattern characteristic of the alert state. However, after a time the subject will become accustomed to the stimulus and the alpha pattern will reappear. The alpha patterns of volunteers given ACTH 4-10 returned more slowly than did those of individuals receiving the control solution. The results of reversal experiments with humans were also consistent with the hypothesis that ACTH 4-10 affects attention.

There have been suggestions that these pituitary peptides are involved in the development of tolerance to morphine. Toler-

Observer's Report

Frontiers of Astronomy:

Sir Fred Hoyle celebrated his 60th birthday this year, and a symposium was held to mark the occasion in Venice, 15 to 19 July. Many of Hoyle's colleagues and former students presented current work in some of the fields Hoyle helped to found, including cosmology, the nature of the interstellar medium, and the early history of the solar system. The following is a report from a participant.

There is no longer really anything to be said about the original steady-state picture of the universe, in which matter is continuously created at just the right rate to keep all average properties constant for all time, except that it is a beautiful idea which turned out not to agree with observations. Hoyle has recently suggested an alternative picture, in which our universe changes with time, not because it is expanding, but because the masses of the stable fundamental particles are increasing with time. Our universe is then just one of many patches in a space-time continuum, each with its own properties, which may have started its life with very large place-to-place fluctuations in the density of matter. S. E. Woosley (Caltech) has calculated the nuclear reactions that should occur early in such a universe, and finds that a large fraction of the material in the densest clumps should be converted to elements with atomic numbers near that of iron. Conventional big-bang universes produce only hydrogen and helium early in their history. Besides providing an additional site for the synthesis of heavy elements, this picture has the attractive property that if the center of our sun is made of material processed in this way, the discrepancy between predicted and observed fluxes of solar neutrinos largely goes away. The chief difficulty may well be to avoid having all the iron so produced get trapped in black holes.

In more conventional cosmology, two of the classic tests, which it was once hoped would tell us whether our universe will expand forever or turn around and recontract, now no longer seem able to tell us this. The first of these tests is the relation between the red shift and the apparent brightness of galaxies. Since the apparent brightness of a galaxy decreases with its distance from us, and distant objects are seen as they were in the past, observations of the recession velocity (red shift) versus apparent brightness should tell us whether the expansion of the universe is slowing down enough for it eventually to stop and begin to contract. The number of available red shifts has increased considerably in the past few years (particularly due to the work of Oke and Gunn at Hale Observatories), but the interpretation has become more difficult. In order to determine the distance to a galaxy from its apparent brightness, we need to know how its real brightness changes with time. It was once assumed that such changes were small, but this no longer seems to be the case. On the one hand, B. Tinsley (Yale University) has shown that the general tendency of the aging of the stars in bright, massive galaxies is to make

ance is the phenomenon in which an individual becomes less sensitive to a drug and requires progressively higher doses of it to achieve the desired effect. William Krivoy of the Addiction Research Center in Lexington, Kentucky, has shown that ACTH and MSH antagonize the analgesic effects of morphine, and that a vasopressin analog facilitates development of resistance to the drug. Investigators have suggested that development of tolerance to morphine is a form of learning and that it involves protein synthesis. Since the hormones are peptides, if not proteins, and are involved in learning, Krivoy has hypothesized that they may somehow participate in the development of morphine tolerance. However, all of the hormones are known to stimulate adenylate cyclase in certain cells. This enzyme catalyzes the synthesis of adenosine 3′, 5′-monophosphate (cyclic AMP). Morphine inhibits the enzyme in cultured nerve cells (*Science*, 29 August). Consequently, the peptides may antagonize morphine activity by counteracting its effects on adenylate cyclase.

Investigators are now trying to identify the neurochemical changes that presumably underlie the effects of the peptides on behavior. De Wied points out that stimulation of adenylate cyclase is one possibility. The known effects of increased cyclic AMP concentrations include changes in membrane permeability, in enzyme activity, and in protein synthesis. Any or all of these could alter neuronal functions and the formation of new connections between neurons.

According to de Wied, ACTH 4-10 does alter protein synthesis. Removal of the pituitary gland decreases the incorporation of the amino acid leucine into protein in the brainstem. The analog of ACTH 4-10 that speeds up extinction of conditioned responses lowers the incorporation even further. But ACTH 4-10 itself restores it to normal. Several investigators have evidence that formation of memory requires protein synthesis. Finally, the idea that the peptide acts through protein synthesis is consistent with the finding that its effects last for several hours even though it is destroyed within minutes.

Studies of the effects of ACTH and MSH on the electrical changes in neurons that occur during transmission of nerve impulses indicate that the hormones increase the excitability of nerve cells. For example, Fleur Strand of New York University showed that ACTH and ACTH 4-10 increase the excitability of peripheral nerve cells and decrease their fatigue during electrical stimulation. And Krivoy, working with Roger Guillemin, who is now at the Salk Institute, showed that MSH increased the excitability of neurons in the spinal cord of cats.

These investigators suggested that MSH, which has no other known function in mammals, might act as a neurohumor (a substance that transmits nerve impulses from one neuron to another). Kastin has shown that the secretion of MSH is controlled very rigorously by means of two hypothalamic factors, one of which inhibits and the other of which stimulates MSH synthesis. Although he admits that the argument is highly teleological, Kastin thinks that such an elaborate mechanism would not have evolved for a material having no function. Transmitting or modulating nerve impulses would, of course, be a very important function.

There have been criticisms of the idea that pituitary hormones affect brain function. One of them centers on the existence of the blood-brain barrier that prevents many substances—especially proteins and peptides—found in the bloodstream from reaching the central nervous system. These hormones, like all others, are secreted into the bloodstream. But vasopressin actually originates in the hypothalamus, and inves-

In Venice

them grow significantly fainter with time, while on the other, J. P. Ostriker and S. D. Tremaine (Princeton) have pointed out that such galaxies will gobble up material from other, nearby galaxies, which will make them grow brighter with time. It does not, at present, seem possible to decide which of the two effects dominates and thus to interpret the red shift–apparent brightness data.

The other classic test is to measure the apparent (angular) diameter of objects whose real size you think you know, as a function of their distance. Because the structure of space-time (within the framework of general relativity) is determined by the amount of matter present, this test should also distinguish an ever-expanding universe from a recontracting one. The optical data on this problem have always been difficult to interpret, but there had been great hopes for using radio sources. R. Ekers and others at the Westerbork Radio Observatory in the Netherlands have recently measured angular diameters of a large number of faint (hence distant) radio sources. Their data, in combination with previous results, say only that the real sizes and brightnesses of radio sources change with time. Since we have no theory of what the changes ought to be, we cannot now do cosmology with angular diameters.

Hoyle was among the first to predict the composition of the dust grains that pervade interstellar space. In the intervening years, his heretical suggestion (graphite) has become part of the conventional wisdom. N. C. Wickramasinghe (Cardiff) has proposed a new heresy, suggesting that polymers of formaldehyde and similar organic molecules may be important components, responsible for the observed ultraviolet absorption features. Even more controversial is the question of what fraction of the interstellar medium's supply of heavy elements is, in fact, locked up in the grains. Analysis of ultraviolet absorption lines in the spectra of distant stars, obtained with the Copernicus satellite, has normally been thought to imply considerable depletion of heavy elements from the gas onto the grains. G. Steigman (Yale University) now points out, however, that most of the absorption probably takes place in ionized regions around the stars themselves, so that the data tell us very little about the heavy element content of the general interstellar gas.

An increasing body of data may require us to rethink our ideas on the history of the solar system during the period when the planets and meteorites condensed and cooled. The traditional view requires a homogeneous gaseous medium to condense quickly and without outside interference. We now know of a variety of place-to-place variations in the ratios of isotope abundances of various elements in meteorites, which require some modification of this picture. D. D. Clayton (Rice University) has suggested that interstellar grains condense in the immediate vicinity of the supernovae where the heavy elements are made and preserve their identity through the condensation process.—VIRGINIA TRIMBLE

The author is visiting associate professor in the Department of Physics and Astronomy, University of Maryland, College Park 20724.

tigators do have some evidence that the system of blood vessels that carries hypothalamic hormones to the pituitary can also carry pituitary hormones to the brain. Kastin, for example, has found MSH in certain parts of the brain. Some investigators have suggested that the effects of the hormones are pharmacological rather than physiological because relatively large doses must be administered in order to produce an effect. The existence of the blood-brain barrier and the fact that the hormones are rapidly broken down in blood help to account for this need to inject large quantities of the peptides.

In any event, the evidence accumulated thus far has raised the possibility that MSH and ACTH 4-10 may be of use in treatment of conditions in which learning or memory is impaired. These include the memory problems that may accompany old age, the temporary amnesia following electric shock treatments for mental disorders such as depression, and the reduced attention span of children suffering from minimal brain dysfunction or hyperactivity. De Wied has found, for example, that ACTH 4-10 and vasopressin can alleviate the amnesia induced in rats when they are subjected to concentrations of carbon dioxide that cause respiratory arrest. This amnesia is a model for that caused by electric shock.

At present, trials of ACTH 4-10 for therapy of the first two conditions are under way, but it is still too early to judge the effectiveness of the agent. One thing in favor of its use is that no side effects have yet been detected. So far there has been little discussion, publicly at least, of the possibility of using these agents to improve the memories of normal people but it is an idea that cannot help but occur to anyone who has ever forgotten anything.

—JEAN L. MARX

Geothermal Resources: A New Look

A new assessment of U.S. geothermal resources underlines the limited utilization being made of this form of energy compared to its potential. The assessment is contained in a report* prepared by the Geological Survey on the basis of new knowledge about geothermal systems and a tabulation of all known systems. The report concludes that at least 12,000 megawatts of electric generating capacity, more than 15 times the current U.S. geothermal output, could probably be achieved at present prices and with current technology. Nearly 10 times that resource, the report estimates, either remains to be discovered or is known but awaits marginally higher energy prices or improved technology. Large quantities of heat at temperatures too low for power generation but adequate for space heating and some industrial uses are also identified, as are the more speculative but large geopressured resources—including heat, high pressure, and methane—underlying parts of the Gulf Coast.

The estimates appear to be conservative. They do not, for example, include any resources below 3 kilometers, the depth to which geothermal wells have already been drilled. The hot and in some cases still molten rocks below that depth, although they constitute an immense store of heat, were considered as beyond the pale of present technology. Nor do the estimates include the large geothermal deposits in Yellowstone, Mt. Lassen, and other national parks, where exploitation would probably destroy the recreational attractions. Additional reasons for believing that the near-term geothermal potential is at least as large as the Survey estimates can be found in the intense interest in these resources in the private economic sector. A virtual explosion of geothermal exploration and drilling activity by oil companies and others has taken place in the past 2 years, despite a host of delays for drilling permits, a hopelessly snarled program for leasing federal lands, and the lack of tax incentives comparable to those available for oil exploration and production.

Despite the future potential of deep, hot igneous rocks and geopressured zones, the main interest at present is in geothermal deposits in which the heat is transferred by hydrothermal convection. The Survey report lists 290 deposits of this type within the United States, about one-fifth of which appear to have subsurface temperatures above 150°C, high enough to be considered for generation of electricity. Subsurface temperatures are estimated on the basis of silicon dioxide and sodium, potassium, and calcium content in water samples from each deposit; these concentrations are thought to serve as chemical geothermometers that in most instances give minimum estimates of the reservoir temperature. Crude estimates were also made of reservoir volume.

As with mineral resources, much of the geothermal heat seems to be concentrated in a few large deposits. Six deposits (five in California and one in New Mexico), each containing more than 10^{19} calories, constitute a large part of the known high-temperature resource. One extended region, the Bruneau-Grandview area of Idaho, contains by itself a staggering 2.6×10^{20} calories (estimated), more than two-thirds of the known intermediate-temperature resource. Reevaluation of intermediate-temperature deposits with newly developed models that take into account the mixing of cooler surface waters and geothermal waters may uncover more deposits of economic interest, the report suggests.

The ultimate source of the heat in hydrothermal systems is thought by many investigators to be a geologically recent magma chamber, but estimates of the number of such chambers and their heat content is more speculative. The report suggests, nonetheless, that perhaps 2.5×10^{22} calories are stored in such chambers in the United States above 10 kilometers depth. Of this total, about half is probably in molten or partially molten form, with temperatures around 650°C. Tapping this energy, however, will involve drilling into the magma chamber and hence, the report concludes, a considerable advance in drilling technology.

Also uncertain is a quite different type of geothermal resource, the geopressured zones found in sedimentary rocks in an area extending from Texas to Louisiana, both onshore and offshore. Geopressured deposits contain hot water at abnormally high pressures, and, in addition, often contain significant amounts of dissolved natural gas. Thus recovery of heat, of mechanical energy (from the high pressures), and of natural gas is potentially possible. Although the area in which these deposits occur has been extensively explored for oil and gas, drilling into geopressured zones has been avoided because of the difficulty in controlling a high-pressure well. The potential resource, however, is large, capable of sustaining an electric generating capacity of 30,000 to 115,000 megawatts and of producing methane of perhaps equal value. The report concludes that this resource is for the most part economically marginal at present energy prices. Taken in combination, however, U.S. geothermal resources are far from contributing at their true potential.—ALLEN L. HAMMOND

*D. E. White and D. L. Williams, Eds., *Assessment of Geothermal Resources of the United States—1975*, Geological Survey Circular 726, Washington, D.C., 1975.