body, and moreover rebuilt to precisely the same structure, one cannot help but speculate about the controlling mechanism. It is rather a terrifying thought that the whole of the protein in the human body is replaced in roughly 160 days, and at the present time we can only speculate on the mechanism controlling this elaborate resynthesis, where even a single amino-acid must not be out of place if the hormone is to have its activity or the antibody its potency.

It might well be that, when the mechanism of perhaps the most complex problem in human biochemistry is understood, we shall find the answer to many problems, possibly even the mechanisms of senescence and of cancer.

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REGULATION OF SECRETION OF CORTICAL HORMONES*

BY

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There are several reasons why information about the control of the rate of secretion from an endocrine gland may be of interest. One is theoretical: an understanding of the factors regulating glandular activity gives us an insight into the part the gland plays in keeping up the *milieu intérieur*. The others are practical. First, in substitution therapy, such knowledge enables us to adapt the quantity of administered hormone to the requirements of the patient, as these vary with the circumstances. Secondly, in trying, for therapeutic reasons, to enhance or inhibit the action of a normal endocrine gland we have to know the conditions which will produce the desired effects on the gland.

Before discussing the information at present available about the control of adrenocortical activity, let me remind you of two fundamental facts concerning the physiology of the adrenal cortex. First, the gland, though secreting continuously, contains practically no stores of the biologically active hormones (Vogt, 1943), and, if synthesis were stopped while secretion were allowed to proceed at a normal speed, would lose its total hormone content in about 6–12 seconds. This means that synthesis is proceeding without interruption in the living condition, and explains the gland's large blood flow and high oxygen consumption. Secondly, the adrenal cortex has no secretory nerves, so that any nervous control over its activity would have to be exerted indirectly.

Experimental Methods

The knowledge we have of the control of adrenocortical secretion is almost entirely derived from animal experiments. A few introductory words on methods will be necessary. All available methods have their uses and their limitations, and the best picture of the gland's performance will be obtained by combining the information each of those methods can provide.

The oldest method is that which determines the experimental conditions which lead to hypertrophy of the cortex. It is suited to the investigation of all stimuli which can be applied over prolonged periods.

For stimuli of short duration suitable methods are the histological or chemical (Sayers *et al.*, 1945) determination of adrenal lipids, cholesterol, or ascorbic acid. Any acceleration of cortical secretion is reflected by an initial depletion in adrenal lipid or ascorbic acid usually followed by a restoration of the stores. This restoration may occur even if the accelerated secretion is maintained, either by prolonged action of the same stimulus or by a new one. Thus depletion is, as a rule, a sign of increased cortical secretion, while full stores of lipid or of ascorbic acid may be found in the later phases of increased activity as well as in the resting condition. Errors in interpretation may therefore arise if these methods are used in the investigation of prolonged or multiple stimuli.

Finally, cortical secretion may be measured directly by assaying adrenal venous blood for its hormone content (Vogt, 1943). The main use of this method is in acute experiments on anaesthetized animals or in perfused glands.

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It has the advantage of providing at least a rough quantitative estimate of hormone production. Also, the interpretation of the results does not depend on the preceding history of the gland.

The Actions of A.C.T.H.

What have these different methods taught us? From the work on adrenal hypertrophy it has been known for a long time that innumerable conditions, usually but not always of a noxious character, stimulate the adrenal cortex. These conditions of "stress" include the action of infections, toxic substances, hormones, abnormal environmental temperatures, high altitude, exercise, haemorrhage—in other words, conditions which will increase the metabolic demands on either the whole body or some of its organs. The most important feature about this action of stress on the adrenal is that, in the mammal, it is abolished by hypophysectomy. The pituitary hormone released by stress and responsible for the adrenal hypertrophy is the "adrenocorticotrophic hormone" of the anterior lobe, or A.C.T.H. The actions of A.C.T.H. are twofold: (1) it maintains the normal size and structure of the adrenal cortex; and (2) it accelerates cortical secretion whenever stress is applied. This action, as shown on the perfused gland, occurs within a few minutes at the latest, and possibly instantaneously.

Since A.C.T.H. is so important for the activity of the adrenal cortex, we need to know how exclusively adrenal cortical function and adaptation of that function to variations in the requirements of the body depend on the pituitary. Obviously, some cortical function persists in the absence of the anterior lobe, or else hypophysectomy would be fatal.

Sections through the adrenal of hypophysectomized mammals show that the atrophy which follows the operation does not affect all zones of the cortex; the outer zone, or zona glomerulosa, is spared, or is possibly even larger than usual. As it is a sound biological argument that structurally different parts of an organ differ in their function, we may assume that, in the hypophysectomized mammal, certain functions of the adrenal cortex will be better preserved than others. There is indeed much circumstantial evidence that the electrolyte metabolism of the hypophysectomized animal is less impaired than that of the adrenalectomized animal. Carbohydrate, fat, and protein metabolism, however, with either procedure, are affected in about equal degree. Generalizations on this subject are, however, dangerous, as there are great species differences in the hypophysial-adrenal relationship. This is well illustrated by the fact that certain kinds of stress which no longer cause cortical hypertrophy in the hypophysectomized rat continue to do so in the hypophysectomized pigeon (Miller and Riddle, 1942).

The next question is whether this function can still be adjusted to the varying demands of the tissues. In the absence of any nervous innervation, any such regulation of the rate of cortical secretion must be mediated humorally by changes in the composition of the blood.

Blood Constituents Other than A.C.T.H.

Are there any blood constituents, other than A.C.T.H., which modify the rate of adrenal secretion? The perfused isolated adrenal of the dog has been used to investigate this problem. On that preparation a number of physiologically occurring substances were tested for their possible influence on the rate of cortical secretion.

Only two means were found to increase the rate of hormone production of the isolated adrenal preparation. One was the injection of adenosine-triphosphate (A.T.P.) and

related compounds, and the other an increase in plasma potassium. A.T.P., when injected into the perfusion cannula, caused a short outburst of enhanced secretion. We know that substances carrying high-energy phosphate bonds enter into the metabolism of most cells, and experiments on the speed of uptake of ^{32}P (Gemzell, 1948) have shown that the adrenal gland is no exception, and also that its ^{32}P uptake is accelerated by A.C.T.H. The perfusion experiments suggest that these phosphates take part in the synthesis of cortical hormones. Normally they are not carried to the gland by the blood stream but are synthesized *in situ*.

The acceleration of cortical secretion by increasing the potassium content of the perfusing blood was obtained with or without simultaneous lowering of the plasma sodium. The alteration in plasma levels had to be of such a magnitude as would be expected to occur only in adrenalectomized dogs. The present methods of hormone assay are not sensitive enough for a decision whether a similar but smaller acceleration would occur if the changes in plasma electrolytes were within physiological range. If that were so, we would conclude that the regulation of cortical secretion which meets the demands of the electrolyte metabolism is entirely independent of pituitary action. It may, however, be that the direct stimulation of the adrenal cortex by excess potassium is an emergency mechanism reserved for pathological conditions only.

Causes of Changes in Pituitary Activity

We come now to the second part of our considerations. If the lion's share of control of cortical secretion is contributed by the pituitary, what, in turn, causes the necessary changes in pituitary activity? There we are still on insecure ground. We do not know whether it is the same factor which acts on the pituitary in all conditions of stress or whether there are several different factors. Two suggestions have, so far, been made for a possible common denominator of all conditions of stress which release A.C.T.H. The first one makes adrenaline responsible (Long, 1947) and is attractive for two reasons: first, adrenaline is undoubtedly secreted during many conditions of stress; secondly, the effect of adrenaline in accelerating cortical secretion is very striking. Infusions of "physiological" quantities of adrenaline—that is, quantities which may be released *in vivo* through prolonged stimulation of the splanchnic nerves—increase cortical secretion immediately, considerably (by many times the basal value), and for a period long outlasting its vascular effects (Vogt, 1944).

The site of action appears to be complex, an important factor being the release of A.C.T.H. This acceleration of cortical secretion by adrenaline constitutes an indirect control of the central nervous system over the function of the adrenal cortex. Is it all we require in order to explain the release of A.C.T.H. in stress?

Such a hypothesis can easily be put to the test, and experiments show that it is untenable. Two groups of rats are submitted to stress, the adrenals of one group having previously been denervated. Both groups of animals show a cortical response to stress—in other words, release A.C.T.H.—whether the method used for measuring the adrenal response be depletion of cortical lipids (Vogt, 1947) or of adrenal ascorbic acid. With the latter method, which gives quantitative data, we can analyse the matter further. Rats with denervated adrenals show the same fall in adrenal ascorbic acid after an injection of insulin as their normal controls. This shows that adrenaline is not indispensable for a response of the pituitary to stress, though it does not mean that adrenaline

plays no part in that response. On the contrary, the rats with denervated adrenals which have been given insulin in a dose well tolerated by normal rats are severely hypoglycaemic, comatose, or convulsing, so that we are in fact submitting the denervated animals to a much severer test instead of observing them and their controls under comparable conditions of stress. Equal depletion of adrenal ascorbic acid may actually constitute a diminished responsiveness of the denervated rat which is able to mobilize its A.C.T.H. only through the hypoglycaemia and not through the secretion of adrenaline as well. The results obtained on rats with denervated adrenals were confirmed in rats completely deprived of both adrenal medullae.

The second suggestion for a common factor releasing A.C.T.H. under all conditions of stress has been made by Sayers and Sayers (1947). Their hypothesis is as follows: in stress there is accelerated utilization of cortical hormones by the tissues, and therefore a low blood level of these hormones. This low level acts as a stimulus to the pituitary, which releases A.C.T.H. Conversely, output of A.C.T.H. is repressed when the level of circulating cortical hormones is high. This has been proved experimentally (Ingle, 1938; Sayers and Sayers, 1945) by showing that injection of cortical hormone inhibits the secretion of A.C.T.H. during stress. The stimulating effect of lack of hormone is much more difficult to demonstrate and has, so far, not been established.

Though we can be certain, therefore, that increased availability to the tissues of cortical hormones will check adrenocortical activity, we cannot be sure that lack of hormone will increase it by causing secretion of A.C.T.H. At present, however, this view is at least a good working hypothesis. It raises a number of further questions: is a low level of circulating cortical hormones the only stimulus which will induce the pituitary to give off A.C.T.H.? How does the role of adrenaline fit into the picture? Does it, too, like other forms of stress, act by increasing the utilization of cortical hormones in the tissues? Further, where are the receptors sensitive to low levels of circulating cortical hormones? Sayers and Sayers pictured them in the anterior lobe itself. For emotional stimuli, however, and for adrenaline, the work of de Groot and Harris (1950) and of Hume and Wittenstein (1950) suggests receptors in the hypothalamus. Does the whole control of A.C.T.H. secretion lie in the hypothalamus? To all these questions we can only guess the answers. And rather than by speculating, I should like to end by discussing a purely practical problem.

Conclusion

Shortage of cortisone and of A.C.T.H. has prompted the search for a treatment of rheumatoid arthritis which will stimulate the pituitary of the patient to secrete enough A.C.T.H. to obtain clinical improvement comparable to that achieved in the Mayo Clinic by injecting exogenous hormones. Insulin has been repeatedly tried, and, according, for example, to a recent report from the Oxford Regional Rheumatism Research Unit, with encouraging success (Kersley *et al.*, 1950). American authors, however, who gave insulin together with cortisone, had less convincing results (Henderson *et al.*, 1950), and Hench stated in a recent lecture at Edinburgh University that in the Mayo Clinic no benefit had been obtained from the combined use of insulin and cortisone. This apparent disagreement is easily explained in the light of the experiments referred to above, in which injected cortical hormones were shown to inhibit the pituitary response to stress. By giving the insulin along with the cortisone, the physician defeats his own ends, because he prevents the very release of

A.C.T.H. which presumably constitutes the beneficial action of insulin in rheumatoid arthritis. Insulin can be expected to have the desired effect only if allowed to act without a simultaneous check on the action of the anterior lobe. In this instance, the fact that cortisone was probably not available to the British workers has proved to be of unexpected benefit to their patients.

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OBJECTIVE ASSESSMENT OF IMPROVEMENT IN RHEUMATOID ARTHRITIS

BY

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Hench (1949) has shown that most of the manifestations of rheumatoid arthritis are potentially reversible. The introduction of cortisone and A.C.T.H. has further shown that reversal can be induced by these hormones, and that many features of rheumatoid disease can be influenced within a few hours of their administration (Hench *et al.*, 1950). In prolonged and intensive courses these changes can be observed by clinical examination and by simple tests of function as shown originally by Hench and other workers in America and more recently by Copeman *et al.* (1950) in this country; and indeed Hench considers that the patient's own assessment of his degree of improvement is one of the more reliable indices of progress. Subjective improvement and simple clinical examination are, however, both very susceptible to psychological factors, and in less experienced hands these methods have led to many false claims of which those made for deoxycortone acetate and ascorbic acid by Lewin and Wassén (1949), LeVay and Loxton (1950), and others are recent examples. These difficulties have been well illustrated by the work of Quin, Mason, and Knowelden (1950).

It would clearly be advantageous to have methods of studying rheumatoid arthritis which would provide some more sensitive and measurable indices of improvement, and if these methods were delicate enough to give a reliable index of the response to single injections of cortisone or A.C.T.H. they would provide a means of comparing the responses of a variety of unknown substances with known standard active and inert preparations all in the same patient. It would also be possible to study the responses of different patients to a single standard active injection, and to compare the responses in the same patients at different stages of the disease, thus providing much valuable information.