EFFECT OF HYPOTHERMIA ON THE HYPOTHALAMIC-PITUITARY RESPONSE TO STRESS

BY

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While it is now established that the functions of the neurohypophysis are controlled by nuclei in the hypothalamus, the mechanisms that regulate the various activities of the adenohypophysis remain obscure. In spite of its independence of any immediate neural control, the adenohypophysis is still capable of adjusting its activities promptly to the varying demands of the body. The almost immediate mobilization of endogenous A.C.T.H. following minor stress stimuli (Sayers and Sayers, 1947) suggests that a reflex mechanism is playing a part in this response. Further evidence for this hypothesis was recently recorded by Porter (1953), who observed that in cats lesions in the posterior hypothalamus block the pituitary-adrenocortical response to stress. When the lesions were made more anteriorly they were ineffective. Porter's observations show that certain functions of the adenohypophysis may also be mediated through the hypothalamic nuclei. Yet despite the many studies the pathway(s) taken by the agent(s) that regulate the activities of the adenohypophysis is (are) still unknown.

The present experiments were carried out to examine the effect of marked lowering of the body temperature on the pituitary-adrenocortical response to stress stimuli. Such hypothermia might selectively depress any neural participation in the release of A.C.T.H. by the adenohypophysis. Evidence is given below to show that with a degree of hypothermia of about 20° C. the adrenal cortex remains responsive to parenteral administration of small doses of A.C.T.H.

Materials and Methods

Sixty-eight male albino rats of inbred Wistar strain and weighing 80–100 g. were used. After keeping them in a room thermostatically controlled at 24° C. for several days they were divided into five groups and treated as follows.

Group I.—After the left adrenal gland had been removed under ether anaesthesia the animals were allowed to recover from its effects for twenty minutes. They were then transferred to a cold room at 0° C. and kept there for thirty minutes. On their return to the laboratory they were re-anaesthetized, their rectal temperature recorded, and preparations made for the removal of the remaining adrenal, which was excised sixty minutes after the first.

Group II.—These rats were treated in the same way as those in Group I except that they were not transferred to the cold room between the two operations.

Group III.—These rats were treated in the same way as those in Group II except that the anaesthesia was maintained for the whole sixty minutes between the two operations.

Groups IV and V were taken in batches of six at a time. They were rapidly anaesthetized with ether and transferred to the cold room. Open ether anaesthesia was maintained during the period of chilling. The rectal temperature of each animal was recorded throughout the experiment. While

in the cold room the anaesthetized animals were unable to maintain their body temperature, which fell rapidly in twenty-five to thirty minutes to about 20° C. (animals that fell to lower temperatures were discarded). They were then returned to the laboratory and placed prone on a grid 1 in. (2.5 cm.) above a large block of ice. The anaesthesia was maintained by occasional open ether. The rectal temperature was maintained at 20–21° C. throughout the experimental period. The respiration rate ranged between 20 and 30, and the heart rate was about 120 a minute. Left adrenalectomy was performed forty minutes after starting the anaesthesia, when the animals were hypothermic. This unilateral extirpation was immediately followed by the injection of adrenocorticotrophin (A.C.T.H., Armour, K55908) in N/30 acetic acid into the tail vein of Group V animals. Doses of 1/500 and 1/250 I.U. were administered to Groups Va and Vb respectively. The same volume of N/30 acetic acid was injected into Group IV animals. The remaining adrenal gland was removed sixty minutes after the first.

Each adrenal gland was dissected free from fat and weighed to the nearest 0.1 mg. Its ascorbic acid content was estimated by the method of Roe and Kuether (1943) as modified by Sayers *et al.* (1948). Colorimetric measurements were made with a Spekker absorptiometer.

Results

The response of these animals to the stresses of anaesthesia, adrenalectomy, and chilling was judged by the degree of ascorbic acid depletion of the second of their adrenal glands; each animal thus acts as its own control. The comparative effects of each of these stressing agents are shown in Table I, from which the following conclusions

TABLE I.—Comparative Effects of the Stress Induced by Various Combinations of Adrenalectomy, Anaesthesia, and Chilling

Group	No. of Animals	Treatment	Mean Rectal Temperature	Ascorbic Acid Content (mg. per 100 g. of Fresh Tissue)		
				1st Gland	2nd Gland	Difference
I	10	Adrenalectomy	33° C.	440 ± 18	230 ± 13	–210 ± 22
II	8	+chilling	35° C.	447 ± 15	272 ± 16	–175 ± 17
III	8	Adrenalectomy + Anaesthesia	33° C.	410 ± 19	230 ± 12	–180 ± 22
IV	22	+adrenalectomy* Chilling under anaesthesia prior to adrenalectomy*	20.5° C.	390 ± 10	396 ± 10	+6 ± 14

* Anaesthesia is mentioned only when maintained throughout the experiment.

can be drawn : (a) That in Groups I and II the operation alone brought about a great and significant fall in the ascorbic acid content of the second gland. This fall was rendered even more severe by the added stress of chilling. (b) That the similarity in the fall in the ascorbic acid content in Groups II and III shows that the maintenance of

TABLE II.—Response of the Adrenal Cortex in Hypothermia to the Intravenous Injection of A.C.T.H.

Group	No. of Animals	Stress	A.C.T.H.	Mean Rectal Temperature	Ascorbic Acid Content (mg. per 100 g. of Fresh Tissue)		
					1st Gland	2nd Gland	Difference
IV	22	Chilling under anaesthesia prior to adrenalectomy*	—	20.5° C.	390 ± 10	396 ± 10	+6 ± 14
Va	13		1/500 I.U.	20.5° C.	370 ± 13	318 ± 14	–52 ± 8
Vb	5		1/250 "	20.5° C.	392 ± 29	323 ± 26	–69 ± 8

* Anaesthesia is mentioned only when maintained throughout the experiment.

anaesthesia throughout the experiment does not mitigate the pituitary-adrenocortical response to the operative stress. (c) That a comparison of the very different changes in the ascorbic acid content in Group IV and the other three groups shows the extent to which the pituitary-adrenocortical response is inhibited by hypothermia.

Further experiments were carried out to determine the degree of responsiveness of the adrenal cortex in hypothermia to varying doses of A.C.T.H. These results are shown in Table II. It will be seen that with a degree of hypothermia of about 20° C. the parenchymatous cells of the adrenal cortex are still sensitive to small doses of A.C.T.H., though there is no linear relationship between dosage and loss of ascorbic acid. Moreover, the response in hypothermia is sluggish when compared with that of hypophysectomized rats at normal temperatures.

Discussion

The rapid fall in the ascorbic acid content in the adrenal glands of animals exposed to various kinds of stress has led to its use for quantitative biological assay of adrenocorticotrophic activity (Sayers *et al.*, 1948). Though the significance of the disappearance of this vitamin is little understood, its loss has been widely recognized as a satisfactory quantitative assay method for A.C.T.H.

The fall in the ascorbic acid concentration in the second adrenal gland in Groups I, II, and III was the expected response of these animals to the various combinations of the stresses imposed by anaesthesia, adrenalectomy, and exposure to cold. When the stresses were imposed on animals that were already hypothermic, as in Group IV, they failed to show any depletion in the ascorbic acid content of the second gland. This implies that at this lowered body temperature the pituitary-adrenocortical mechanism was inhibited. That this inhibition was not due to the effect of the maintained anaesthesia is shown by the response of Group III animals, which were kept anaesthetized throughout the experiment. Nor could it be attributed wholly to lack of sensitivity on the part of the adrenal cortex, for Group V animals showed that with this degree of hypothermia the adrenal cortex still shows some response to small doses of A.C.T.H.

It can be concluded from these findings that at this degree of hypothermia the adeno-hypophysis does not release any appreciable amount of A.C.T.H. in response to the stresses applied. Whether this block is due to an inhibition of certain metabolic activities in the adeno-hypophysis or to the interruption of a neural or neuro-humoral phase in the response mechanism is not clear from the present experiments. The evidence of hypothalamic involvement in the pituitary-adrenocortical response to stress (Porter, 1953) favours the view that, in hypothermia, an inhibition of neural transmission of stress stimuli is mainly responsible for this block. The study of the effects of hypothermia on the stress response may be a useful guide to the further understanding of the mechanism involved.

Summary

Evidence is given to show that in hypothermic rats the hypothalamic-pituitary mechanism leading to the release of A.C.T.H. in response to stress stimuli is inhibited. The adrenal cortex in such chilled animals, however, remains responsive to the intravenous injection of small doses of A.C.T.H.

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A SIMPLE METHOD OF RAISING AND LOWERING BODY TEMPERATURE

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While studying the effects of stress in hypothermic rats it was noted that at a body temperature of 20° C. the pituitary response was inhibited and that of the adrenal cortex was much reduced (Khalil, 1954). With these small animals changes in body temperature could be effected readily; when dogs and rabbits have been made hypothermic, rewarming by the external application of heat takes much longer than with rats, and this delay may have contributed to the death of some of the animals. The introduction of warm water into the stomach could provide a simple and rapid method for supplying heat to the body, since the stomach wall is highly vascularized and lies close to the aorta, vena cava, portal vein, and other large vessels. Conversely, by using cold water a marked degree of hypothermia can be attained.

Since repeated changes of cold or warm water are needed to effect the desired temperature changes, the water is introduced through a tube which joins a rubber balloon lying in the lumen of the stomach. By this procedure the water remains "external" to the body and any loss of electrolytes by gastric lavage or excessive absorption of water is avoided.

The apparatus used consisted of a thin "portex" plastic tube with a finger-stall, which on inflation could readily hold 250 ml., tied securely to one end. The other end was attached to a two-way tap connected with a 50-ml. syringe and with a rubber inlet tube from the warm or cold water to be used. The temperature of the former is about 40° C. and of the latter about 4° C. The irrigation was carried out once every five minutes, each change of water remaining in the balloon for about three minutes. The method was employed for rabbits and was effective in lowering the rectal temperature to about 25° C. in two hours and to 20° C. in three hours and in raising it to normal again in about four hours. The heart rate was slowed from 240 to 60 beats per minute. This work will be reported more fully later.

At this stage an opportunity arose of using this method for cooling a 16-months-old child with hyperpyrexia who had not responded to the usual methods of cooling.

Case History

A boy aged 16 months was admitted to hospital on June 29, 1954, with pneumonia, which resolved with treatment. There was a family history of fits, and the boy had had severe fits at 10 months while teething. On the first and second day after admission he had fits and was left with a right hemiparesis and regression in general behaviour to that of a 10-months-old child, although he was alert and active. On July 15 he was transferred to Guy's Hospital, and on July 16 an arteriogram was done which showed slight shift of cerebral vessels to the left. On the morning of July 20 he had a temperature of 104° F. (40° C.); no abnormality was found in the throat, ears, lungs, and urine. At 1 p.m. he was stuporous and very ill, with cold skin