Contribution to Understanding of Mechanism of Permissive Action of Corticoids. (23022)

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The mechanism of interaction in metabolism between corticoids and response to injury, has been the subject of considerable debate in recent years(1,2,3). One issue has been whether corticoids should be considered as "prime movers" in the response or whether their role is important but basically secondary one. The first point of view is expressed clearly in the following interpretation of the hyperglycemic response to trauma written by Selye and Horava in 1953(4), and describing the results of an earlier experiment of Selye and Dosne(5). "Traumatic shock produces hyperglycemia in the intact, but hypoglycemia in the adrenalectomized rat. If an adrenalectomized rat is maintained on threshold doses of adrenocortical extracts (which in themselves cause no hyperglycemia), it can again respond to surgical shock normally, that is by a marked rise in blood sugar. Apparently the stress of traumatic injury does not merely raise the blood sugar through excessive production of glucocorticoids, nor do the glucocorticoids exert their maximal hyperglycemia action by themselves. We may conclude that the metabolic changes incident to stress produce favorable conditions for the manifestation of the hyperglycemic effect of the glucocorticoids." Although Selye qualifies this interpretation by writing " . . . it is immaterial whether we say that the hormone conditions the effect of the non-hormonal stimulus, or vice versa, since synergism between both factors is necessary to obtain an optimal response," it is still of considerable importance for the interpretation of the theory of diseases of adaptation to establish the order of causality, if possible. This is particularly true since Selye has written elsewhere(1). "Stress itself is perhaps the most effective and most common factor capable of conditioning the action of adaptive hormones." If trauma conditions the action of the glucocorticoids then the hormone must be considered the cause of the response. Extending this to Selye's concept of the diseases of adaptation the hormones become the chief mediators in some of these diseases which in turn must be considered as examples of hypercorticalism. The alternate point of view, championed by Ingle(2) and favored by ourselves(3), is that hormones play a supporting role in the reactions to trauma. In these terms the corticoids make conditions suitable for a normal response to trauma and as such presumably are tending to subserve homeostasis. The role of hormones in the etiology of diseases would then be considered as secondary to other factors. Thus, they might provide conditions suitable for development or manifestation of disease or might accentuate the signs or symptoms of disease but not themselves cause the disease. The activation of a peptic ulcer by glucocorticoids(6) may be an example of the first mechanism while accentuation of edema by aldosterone in nephrosis and congestive heart failure may be an example of the second. Since it has been shown that hyperglycemia of trauma depends chiefly on hepatic glycogenolysis(7), it has always seemed more reasonable to us to assume that the absence of this hyperglycemic response to trauma in the adrenalectomized rat relates in part at least to deficient stores of liver glycogen in the fasted adrenalectomized rat. If so, administration of cortical extract in Selye and Dosne's experiment would permit hyperglycemia simply by promoting gluconeogenesis and glycogen formation, thereby providing an hepatic source of glucose. If this interpretation is correct, one would anticipate that the fed adrenalectomized rat would exhibit an hyperglycemic response to trauma since it is well known that this preparation has adequate liver glycogen stores.

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TABLE I. Influence of Trauma on Blood Glucose-Adrenalectomized Rats.

Experimental groups	No. of rats	Blood glucose, mg % (mean ± S.E.) 0 hr	1 hr	2 hr	3 hr
Control					
1. Adrx.–fasted–saline–DOCA	6	51 ± 3.7	57 ± 6.6	58 ± 4.3	51 ± 5.3
2. " –fed–saline–glucose–DOCA	5	110 ± 12.0	104 ± 8.7	96 ± 5.0	85 ± 4.8
3. " –fasted–saline–DOCA–cortisone	5	119 ± 10.6	108 ± 6.2	103 ± 7.5	92 ± 8.7
Traumatized					
4. Normal–fasted	6	79 ± 4.4	157 ± 12.4	153 ± 15.8	142 ± 14.2
5. Adrx.–fasted–saline–DOCA	11	62 ± 4.7	69 ± 5.2	59 ± 8.6	55 ± 8.6
6. " –fed–saline–glucose–DOCA	6	117 ± 4.5	151 ± 10.8	131 ± 13.1	125 ± 14.8
7. " –fasted–saline–DOCA–cortisone	6	69 ± 3.2	105 ± 7.8	90 ± 7.6	82 ± 11.5

The present experiment demonstrates the validity of this hypothesis.

Method and materials. Male albino rats of the Vanderbilt strain, weighing between 150 and 250 g were used. The animals were anesthetized with pentobarbital and bilaterally adrenalectomized through lumbar incisions. The following groups were studied. (1) Normal rats, fasted 16 hours. (2) Adrenalectomized rats maintained with 0.9% saline in their drinking bottles and subcutaneous injections of 0.5 mg DOCA daily. They were fasted 16 hours before the experiment. (3) Adrenalectomized rats as in group (2) but allowed free access to Purina Chow and 2.5% glucose in 0.9% saline until the time of trauma. To insure adequate liver glycogen stores, the animals were tube-fed 5 ml of a high carbohydrate liquid diet(8), 5 hours before trauma. (4) Adrenalectomized rats maintained with 0.5 mg DOCA and 1 mg cortisone acetate i. m. daily, 0.9% saline and fasted 16 hours. All animals were allowed to drink freely up to the time of trauma. The rats, anesthetized with pentobarbital, were traumatized 3-5 days after adrenalectomy in the same fashion as described by Selye and Dosne. *i.e.* by crushing the abdominal muscles, the stomach and cecum with a hemostat. Blood was drawn from the tail prior to trauma and 1, 2 and 3 hours thereafter, or at the corresponding times in the controls, and were analyzed for glucose by the method of Somogyi(9).

Results. The results are summarized in Table I. As expected, the fed and cortisone treated fasted adrenalectomized rats had significantly higher blood sugar levels than the fasted adrenalectomized animals during periods of observation without trauma. After trauma the normal fasted rat had an hyperglycemic response which the untreated fasted adrenalectomized animal did not share. In contrast to Selye and Dosne's results, however, the fasted adrenalectomized rats as a group did not develop significant hypoglycemia, although individual animals did. The difference in response is probably due to the fact that the rats in Selye and Dosne's experiments received only tap water to drink during the 16 hours fast, whereas our rats were protected somewhat against vascular collapse by DOCA and saline. The vascular collapse is a potent factor in causing hypoglycemia(7).

When the adrenalectomized rats were prefed carbohydrate, they exhibited blood sugar changes in response to trauma which were not significantly different from those of normal fasted traumatized rats. Finally, the adrenalectomized fasted group which had been pretreated with cortisone had an increase in blood sugar after injury which was of the same magnitude as the fed adrenalectomized animals, but was less sustained.

Discussion. These results demonstrate clearly that cortical hormone is not necessary for the hyperglycemic response to trauma, provided a source of glucose is present, presumably as liver glycogen. They make unnecessary Selye's assumption that trauma conditions the actions of the corticoids and hence that the hyperglycemia of trauma is a manifestation of hyper-

adrenalcorticism. In point of fact, they are more consistent with the reasoning used in Selye and Dosne's original report, in which the hyperglycemia was attributed to a continuing ability to resynthesize glucose from lactic acid in the corticoid treated rats, even though this is probably not the precise mechanism involved. They also conform better with Selye's earlier discussion in *Stress*(10) where the significance of glycogen stores for the hyperglycemic response to trauma is alluded to. In light of these considerations, Selye's more recent interpretation of these experiments is puzzling.

The demonstration that either the fed or the cortisone maintained fasted adrenalectomized rat reacts to trauma with hyperglycemia fits well with Ingle's "permissive" concept. In a real sense, cortical hormone may be considered as "permitting" the hyperglycemic response to trauma by making conditions metabolically favorable for a continuing supply of glycogen in the liver and consequently blood glucose. The same end is met by feeding. Obviously, in this example, neither adrenal hormone nor feeding *cause* the hyperglycemia of trauma, even though each by itself may raise the blood sugar if given in excess. Both simply provide the conditions which are necessary for the trauma to initiate an hyperglycemic response even though they do so through different primary mechanisms. This experiment illustrates in a graphic way what we understand by Ingle's "permissive" concept. The same formulation is unquestionably applicable to many other aspects of the adrenal-stress interaction, but not necessarily to all of them. The permissive concept does not imply an "all-or-none" effect as Selye(1) has repeatedly, but incorrectly, attempted to read into Ingle's use of the word "permissive" by the use of a light-switch analogy. As illustrated in the present report in some cases the hormone as well as other factors may "permit" the reaction to go on, while in other reactions the hormone may be essential and not replaceable by some other factor. Moreover, the magnitude of the response may be influenced by the hormone without it being necessary to assume that the hormone is the prime mover. Thus, in the present experiment, the dose of hormone would determine the level of the liver glycogen but the latter is reflected in only a modest elevation in blood sugar unless some other factor, such as trauma (as in this experiment), pancreatic insufficiency or something else enters the picture. The term "permissive" describes this type of biological interrelationship very well.

Selye has applied "conditioning" to this interaction but he has used and defined this term so variably in different publications that it now has a less specific meaning than "permissive" as used by Ingle and ourselves. Thus, within one year the following definitions of "conditioning" and "conditioning factors" may be found in Selye's writings: (1) "Conditioning factors are factors which have little or no activity but can significantly alter a response to a stimulus (*e.g.* sodium can act as a conditioning factor of DOCA activity)"(1). (2) Conditioning factors are "substances or circumstances which influence the response to an agent, for instance, a hormone."(11). (3) "Conditioning is a generic designation to include synergism, antagonism, potentiation, induction of reactivity where such would otherwise not exist (Ingle's original meaning of permissive)† and qualitative changes in reactivity"(12). These definitions are so broad that they tend to obscure rather than clarify our understanding of the mechanism of interaction of the hormones in the stress response.

We believe that the permissive concept as used by Ingle and defined and illustrated above, serves as a useful working basis not only for the understanding of the metabolic phenomena of stress just discussed, but also for the elucidation of the role of the hormones in certain pathological processes such as peptic ulcer, hypertension, inflammation, etc. We have discussed the application of this concept to these problems elsewhere(6,13). When more information is available concerning the mechanisms involved in each of the

† It should be noted that Ingle has never defined "permissive" in this manner. The above is Selye's misinterpretation of Ingle's use of the term.

reactions under consideration descriptive terms such as "permissive" and "conditioning" will have outlived their usefulness. Indeed, the continued use of these terms without acknowledgment of mechanisms when they are known serves to confound rather than clarify our understanding. An example of this may be found in a recent report(14) which refers to a high sodium intake as conditioning the action of desoxycorticosterone to cause muscular paralysis in dogs. Nowhere does the author acknowledge the well established facts that the paralysis is due to potassium depletion secondary to the influence of DOCA on renal excretion of potassium and that a high sodium intake accentuates potassium depletion by promoting renal loss of potassium.

Summary. The adrenalectomized rat may exhibit a normal hyperglycemic response to trauma if maintained with a constant dose of glucocorticoids while fasting or if untreated but fed. In both cases the normal response is interpreted as being dependent on an availability of liver glycogen to permit an hyperglycemic response while the failure of the fasted untreated adrenalectomized rat to develop hyperglycemia may be due to deficient glycogen stores rather than to hormone lack *per se*. The significance of these findings is discussed in relation to current concepts of the interaction of the adrenal cortex and the stress reaction.

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Influence of Parathyroids on Removal of Citric Acid Administered by Peritoneal Lavage.* (23023)

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Since Dickens(1) discovered that bone contains large quantities of citric acid, there has been much interest in the relationship of this acid to various bone constituents, and in its relation to over-all bone metabolism. Carlsson and Hollunger(2) have shown that citric acid may be produced in bone and reported an increased production of this acid following administration of vit. D. Elliott and Freeman(3,4) reported that citric acid changes parallel those of calcium in plasma of various animals and suggested that plasma increases in citric acid as well as those in calcium were probably supplied by bone. They further noted that either vit D or parathyroid hormone were able to raise plasma levels of both calcium and citrate. Neuman *et al.*(5) reported their belief that bone produces citric acid in response to parathyroid hormone which in turn is complexed with calcium and released into the plasma. It has also been shown(6) that endogenous citric acid, pro-

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