THE MAJORITY of living forms depend for their functioning upon two classes of biocatalysts, the enzymes and the hormones. These biocatalysts permit the diverse chemical reactions of the organism to proceed at 38°C with a specificity and at rates frequently unattainable in vitro at elevated temperatures with similar reactants.

The physiologic importance of enzymes and hormones is evident not only under normal circumstances, but is reflected clinically in the diverse descriptions of errors of metabolism, due to lack or deficiency of one or more enzymes, and the numerous hyper- and hyperfunctioning states resulting from imbalance of hormonal supply.

Inasmuch as both enzymes and hormones function, with rare exception, to accelerate the rates of processes in cells, investigators have sought possible interrelationships and interactions of enzymes and hormones, particularly as a basis for the mechanism of hormonal action. It has seemed logical to hypothesize that hormones, while not essential for reactions to proceed but nevertheless affecting the rates of reactions, may function by altering either the concentration or activity of the prime cellular catalysts, the enzymes. This proposed influence of hormones on enzymic activity might be a primary, direct effect achieved by the hormone participating as an integral part of an enzyme system, or an indirect influence based upon the hormone altering the concentration of available enzyme and/or substrate utilized by a particular enzyme.

It is the purpose of this presentation to describe a relatively few, but better defined, examples of the more direct relationships of enzymes and hormones. Five examples of enzyme-hormone interaction will be presented, based on the criterion that an effect of the hormone has been demonstrated on addition of the hormone in vitro to a purified, or partially purified, enzyme system. Other additional examples are available in the literature, e.g., the studies\(^1\)\(^2\) describing the influence of deoxycorticosterone and other steroids added in vitro on the activity of a variety of enzyme systems. It is also true that many publications have described alterations in the activity of enzymes in various tissues following administration in vivo of diverse hormonal preparations. However, it is not possible to judge, in the in vivo experiments, whether the reported effects are examples of direct enzyme-hormone interaction, or an indirect influence of the hormone mediated via one or more metabolic pathways, and therefore other enzyme systems whose activities are not being measured. Data from in-vivo studies of this type are thus not pertinent to a discussion of direct hormone-enzyme interaction.

Before considering the main topic of direct enzyme-hormone interaction, it might be well to recall another aspect of enzyme-hormone interaction which is of profound significance for the organism, viz., the alteration of hormonal structure and activity produced by a variety of specific enzyme systems. This information may belong correctly to the subject of hormone metabolism rather than under the topic of possible metabolic influences of hormones at an enzymic level, with which we are concerned in the present discussion. Nevertheless, enzyme-hormone interaction, as seen, for example, in the profound role of the enzymes of the...
liver in the metabolism of certain hormones, is of paramount importance in determining the effectiveness of these hormones. The ability of the organic chemist to prepare synthetic hormonal derivatives which are relatively resistant to enzymic processes in the liver has been of outstanding value for approaches to oral hormonal therapy. Largely unexplored as yet is the possibility that enzyme-hormone interactions may lead to the production of physiologically more active substances from compounds normally synthesized and secreted by a particular endocrine gland.

It may be said at the outset that in no instance has a hormone been demonstrated to influence the rate of a cellular reaction by functioning as a component of an enzyme system. In Table I are listed five examples of hormone-enzyme interaction for which experimental documentation is, at least in part, satisfactory. It will be noted that each of the examples listed indicates an augmenting action of the hormone on the activity of the specific enzyme system affected. In a real sense, therefore, the influence of these particular hormones results in an acceleration of the rate of a specific cellular reaction. Let us now consider briefly each of these examples in Table I.

It may be pointed out initially that in the case of three of the hormones in Table I (adrenocorticotropin, epinephrine and glucagon) a similarity exists in the effects of these hormones in that each produces an increase in phosphorylase activity, albeit in different tissues. The augmented rate of re-phosphorylation of inactivated phosphorylase, produced by epinephrine and glucagon via their action on the dephosphophosphorylase kinase enzyme system, results in augmentation of the activity of phosphorylase. Whether adrenocorticotropin acts on a similar enzyme system in the adrenal gland is presently unsettled. In any case, adrenocorticotropin augments adrenal phosphorylase activity, epinephrine increases phosphorylase activity in both liver and muscle, while glucagon elevates phosphorylase functioning solely in the liver. What is of considerable interest is the evidence\(^3,4\) that each of these three hormones produces its effect on the phosphorylase systems by promoting the synthesis of an anhydride of adenylic acid, adenosine 3',5'-monophosphate. The reaction catalyzed by each of the three hormones is the following:

\[
\text{Adenosine triphosphate (ATP)} \xrightarrow{\text{adenosine triphosphatase}} \text{adenosine diphosphate (ADP)} \xrightarrow{\text{adenosine 3',5'-monophosphate (AMP)}}
\]

The adenosine 3',5'-monophosphate functions as a cofactor in the enzyme system concerned with the activation of phosphorylase; in the case of liver and muscle, this is the dephosphophosphorylase kinase system. Thus epinephrine and glucagon function to activate phosphorylase, thereby promoting the conversion of glycogen to glucose 1-phosphate, leading to augmentation of the level of glucose in the blood. In the adrenal cortex, it has been hypothesized that an increased activity of adrenal phosphorylase provides additional glucose 6-phosphate,
from which reduced triphosphopyridine nucleotide can be obtained as a consequence of the operation of the phosphoglucuronate oxidative pathway. In turn, the reduced nucleotide participates in adrenal steroid bio-
genesis, notably in the hydroxylation mechanisms which lead to introduction of hydroxyl groups into the steroid nucleus. Haynes and his collaborators have recently made the striking observation that the previously named anhydride of adenylic acid functions more efficiently than does adrenocorticotropic in stimulating corticosteroid synthesis in vitro by slices of rat adrenal gland. Thus, although there are marked differences in the end result achieved by adrenocorticotropic, on the one hand, and epinephrine and glucagon, on the other, as a result of their augmentation of the phosphohexylase reaction, it is nevertheless of considerable interest that each hormone (a chemically distinct substance from the others) has as a basis for its action at the enzymic level a stimulation of the synthesis of adenosine 3',5'-monophosphate. The mechanism by which these hormones promote this synthesis is at present unknown.

The role of estradiol in augmenting the catalytic activity of an enzyme system present in human placenta, endometrium, mammary gland and the pituitary has been described. Detailed studies of this system have been conducted with preparations derived from placenta; it has been demonstrated that the estrogen-sensitive system catalyzes a pyridine nucleotide transhydrogenation:

\[
\text{TPNH} + \text{DPN}^+ \rightarrow \text{DPNH} + \text{TPN}^+.
\]

The group of enzymes catalyzing this reaction has been studied extensively and termed pyridine nucleotide transhydrogenases. It has been proposed that the estrogen activates in some unknown manner a specific, soluble placental pyridine nucleotide transhydrogenase. On the other hand, it has been observed that a variety of 3α-hydroxysteroid dehydrogenases, as well as estradiol-17β-dehydrogenase, could function as transhydrogenases. From this evidence, it was hypothesized that estrogens may function physiologically as coenzymes or cosubstrates in transhydrogenase reactions. For example, by alternately oxidizing and reducing estradiol-17β, the specific dehydrogenase, utilizing different pyridine nucleotides (DPN and TPN) for the two reactions, could effect a net transfer of hydro-
gen from one nucleotide to another. The hormone thus was visualized as serving as a coenzymic hydrogen carrier. However, it has been reported that the mitochondrial transhydrogenases, in contrast to the soluble fractions of human placenta and rat liver, are unaffected in activity by additions of steroid. Moreover, the very low absolute activity of the transhydrogenase reaction catalyzed by hydroxysteroids, and the relatively low activity of the dehydrogenase reaction as compared to the transhydrogenase reaction, make it unlikely that such low activities could be of importance in regulat-
gening the flow of electrons between the two forms of the pyridine coenzymes, even in the soluble portion of the cell. It may also be pointed out that it has recently been demonstrated that the estrogen-sensitive system of human placenta is a soluble pyridine nucleotide transhydrogenase, separate and distinct from the estradiol dehydrogenase present in the same tissue. It would appear, therefore, that it may be premature to conclude that the physiologic basis for estrogen action, at the enzymic level, has been established.

The fifth and final example of hormone-enzyme interaction to be presented has been studied recently in our laboratory. Certain adrenal cortical steroids, notably deoxycorticosterone and cortisol, have been demonstrated to produce in vitro a striking inhibition of glucose utilization by cell-free extracts of normal and malignant lymphoid tissue. This effect has been localized in at least two steps in the over-all oxida-

* TPNH and TPN = reduced and oxidized triphosphopyridine nucleotide, respectively; DPNH and DPN = reduced and oxidized diphosphopyridine nucleotide, respectively.
published data indicate that, under these conditions, an increased phosphate uptake into triosephosphates is counterbalanced to a considerable extent by the release of inorganic phosphate from ATP by the action of the tumor ATPase. In the presence of deoxycorticosterone (Exp. 2b) the release of inorganic phosphate into the medium is increased to such an extent that no net uptake of phosphate is observed; concomitantly, glucose disappearance and lactate accumulation are inhibited.

The deoxycorticosterone and ATPase effects can be overcome almost completely when a system is provided which generates a substrate-level quantity of ATP (Exps. 2c and 2d).

The results thus demonstrate an effect of deoxycorticosterone on a cell-free preparation where permeability factors, at least with respect to cell wall membranes, are not involved. The data suggest a role for the steroid in the reactions of anaerobic glycolysis, particularly in those specific reactions involving the utilization and formation of ATP. In this connection, one group reported inhibition of hexokinase in vitro by cortisol, and another described an inhibition of hexokinase in vitro by deoxycorticosterone. However, we have been unable to confirm the data of either of those studies. It may be noted that there has been described an in-vitro combination between ATP and certain steroids, including deoxycorticosterone. A suggested interpretation of our data is that such complex formation between steroid and ATP could be reflected in an influence of the steroid on certain enzymic reactions which are ATP-dependent.

In concluding, it may seem disappointing, in view of the large body of evidence for the participation of certain of the vitamins as coenzymes in well-described enzyme systems, not to have uncovered a similar broad basic principle which would aid in explanation of the influence of hormones on the rates of cellular reactions. However, a significant beginning has been made in dissecting the mode of action of certain hor-
THE CELL

TABLE II

In-vitro inhibition by deoxycorticosterone (DOC) of anaerobic glucoolysis in cell-free preparations of rat lymphosarcoma

Incubation mixture of 3 ml contains: cell-free preparation from 30 mg tissue, wet wt.; 7.3 µmole potassium phosphate buffer, pH 7.4; 75 µmole KHCO3; 80 µmole MgCl2; 46 µmole KCl; 40 µmole KF; 15 µmole potassium pyruvate; 120 µmole nicotinamide; 0.6 µmole K2DPN; and 10 µmole glucose. Where present, 1.32 µmole DOC. ATP as indicated below. Mixtures incubated for 1 hr at 37.5°C under 95% N2—5% CO2 and then deproteinized with 5% trichloroacetic acid. Lactate, P1, and true glucose determined on trichloroacetic acid filtrates. 3'

<table>
<thead>
<tr>
<th>Exp.</th>
<th>ATP Source</th>
<th>DOC</th>
<th>(µmole per hour)</th>
<th>Δ Lactate</th>
<th>P1 uptake</th>
<th>Δ Glucose</th>
<th>P1</th>
</tr>
</thead>
<tbody>
<tr>
<td>1a</td>
<td>Present initially*</td>
<td>-</td>
<td>4.35</td>
<td>+8.90</td>
<td>2.20</td>
<td>1.1</td>
<td></td>
</tr>
<tr>
<td>1b</td>
<td>Present initially</td>
<td>+</td>
<td>2.48</td>
<td>+0.90</td>
<td>1.00</td>
<td>2.7</td>
<td></td>
</tr>
<tr>
<td>1c</td>
<td>Generated**</td>
<td>-</td>
<td>4.75</td>
<td>+8.80</td>
<td>3.80</td>
<td>1.3</td>
<td></td>
</tr>
<tr>
<td>1d</td>
<td>Generated**</td>
<td>+</td>
<td>2.25</td>
<td>+1.00</td>
<td>0.70</td>
<td>2.3</td>
<td></td>
</tr>
<tr>
<td>2a</td>
<td>Present initially†</td>
<td>-</td>
<td>7.17</td>
<td>+4.38</td>
<td>7.12</td>
<td>1.6</td>
<td></td>
</tr>
<tr>
<td>2b</td>
<td>Present initially†</td>
<td>+</td>
<td>5.34</td>
<td>-8.13</td>
<td>2.59</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>2c</td>
<td>Generated‡</td>
<td>-</td>
<td>6.68</td>
<td>+9.60</td>
<td>5.60</td>
<td>0.7</td>
<td></td>
</tr>
<tr>
<td>2d</td>
<td>Generated‡</td>
<td>+</td>
<td>5.56</td>
<td>+8.70</td>
<td>4.90</td>
<td>0.6</td>
<td></td>
</tr>
</tbody>
</table>

*1.0 µmole K3HATP (catalytic level).
**1.0 µmole ATP generated from 1.0 µmole each of K2HADP and sodium phosphocreatine by 0.3 µg purified transphosphorylase (generously supplied by Dr. Henry A. Lardy).
†10.0 µmole K3HATP (substrate level).
‡10.0 µmole ATP generated from 10.0 µmole each of K2HADP and sodium phosphocreatine by 20 µg transphosphorylase.

Enzymes on enzyme systems. This area of investigation holds promise of rapid expansion in the immediate future. In this regard, a word of caution is perhaps warranted. The precise mechanism of action of hormones, at a cellular level, might not prove to be mediated solely, or perhaps at all, via direct effects on complete enzymic systems. Perhaps more attention should be directed toward an indirect hormonal influence at sites regulating the concentration of components of enzymic systems, that is, the substrates and cofactors made available to the intracellular reactions whose rates are enzymically controlled.

REFERENCES

9. Hagerman, D. D., and Villee, C. A.:


THE INTERACTION OF ENZYMES AND HORMONES
Abraham White

Pediatrics 1960;26:476

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