reaction is similar to raising the ionic strength or lowering the temperature, that is, a disruption of the actomyosin complex occurs.

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Action of Insulin on Thiamine Phosphorylation

Previously we have observed that the respiratory quotient of animals, in which alloxan diabetes reaches a certain severity, is not appreciably affected by administration of thiamine + glucose[1]. Also, under the same experimental conditions, the administration of thiamine was not followed by a marked increase of liver cocarboxylase, whereas in normal animals or in those affected by less severe forms of the disease, such an increase does occur[2]. Therefore it was concluded that in alloxan diabetes a disturbance of the thiamine phosphorylation processes may occur.

In order to investigate whether insulin acts on the formation of cocarboxylase from thiamine or not, we have studied its action on those forms of alloxan diabetes in which the phosphorylation of thiamine is nearly completely inhibited. To such an end we have worked with diabetic rats, severely affected, in which the respiratory quotient was not increased after 1–1½ hr. from the time of glucose + thiamine administration.

In a typical experiment the liver cocarboxylase content of a diabetic rat treated with insulin (20 I.U./kgm.) + thiamine (15 mgm./kgm.) was determined and compared with the liver cocarboxylase content of diabetic rats which had been treated with thiamine alone (15 mgm./kgm.). In all experiments the animals were killed 40 minutes after the corresponding treatment. Cocarboxylase was determined by the method of Ochoa and Peters[3] with minor adaptations[4].

Cocarboxylase Content of Liver of Alloxan Diabetic Rats, Untreated and After Treatment with Thiamine (15 mgm./kgm.), with Insulin (20 I.U./kgm.) and with Thiamine (15 mgm./kgm.) + Insulin (20 I.U./kgm.)

Rats	No. of animals treated	Liver cocarboxylase (μgm./gm. wet tissue)	
		Range	Average
Diabetic	15	3·64–5·20	4·35 ± 0·16
Diabetic + thiamine	15	4·12–6·03	5·04 ± 0·19
Diabetic + insulin	15	3·93–6·17	4·73 ± 0·21
Diabetic + thiamine + insulin	15	6·40–9·49	7·68 ± 0·26

It will be seen from the accompanying table that liver cocarboxylase content is increased in diabetic animals treated with thiamine + insulin, in contrast to those treated with thiamine alone. On the other hand, administration of insulin (20 I.U./kgm.) to diabetic rats not treated with thiamine does not give rise to a significant increase in the cocarboxylase content when compared with the untreated rats. The negative results obtained with insulin alone may be attributed to a low content of the thiamine in diabetic animals[5], probably due to a low absorption of thiamine through the intestine[1]. Hence we may conclude that insulin enhances the thiamine phosphorylation in the diabetic animal when the latter has a sufficient amount of thiamine.

A full report of this work will appear elsewhere.

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Separation of Coombs Reagent into Two Fractions

It was shown by Dacie[1] that normal human sera contain what appears to be an incomplete antibody which becomes attached to erythrocytes at 4° C. and which can be demonstrated by the antiglobulin technique of Coombs. It appears, however, that the substance in the Coombs reagent (rabbit anti-human globulin serum) involved in the reaction is distinct from that involved in the agglutination by Coombs reagent of red cells sensitized by incomplete *Rh* antibodies, since the Coombs reagent can be separated into two fractions by absorption with suitably sensitized cells.

Fraction 1, made by absorbing twice with an equal volume of cells sensitized by Dacie's cold incomplete antibody, agglutinates *Rh*-sensitized cells almost as well as the unabsorbed Coombs reagent while failing to agglutinate cells sensitized by Dacie's antibody.

Fraction 2, made by absorbing three times with an equal volume of *Rh*-sensitized cells, agglutinates to an almost undiminished titre cells sensitized by Dacie's antibody, but gives no reaction with *Rh*-sensitized cells (see Table 1).

Table 1. Absorption of Coombs Reagent with Sensitized Cells

Absorbing cells	Titre: Test cells: *Da*	*Rh*
Nil	16	32
Da	Nil	16
Rh	8	Nil

Da, cells sensitized by Dacie's cold incomplete antibody.
Rh, cells sensitized by incomplete *Rh*-antibody.

That the reaction is of a similar nature to that described by Coombs is shown by the fact that normal rabbit serum does not agglutinate cells sensitized by Dacie's antibody; it is also shown by the inhibition of the reaction by human serum or human gamma-globulin (see Table 2). The titres shown in Table 2 would appear to indicate that two different fractions of human serum are responsible for inhibiting the two reactions, and that the purification of the gamma-