The dispensability of insulin at high sugar concentrations is in agreement with the well established evidence that completely diabetic animals can store some glycogen if high blood sugar levels are maintained.[7, 8]

The relationship between the action of insulin and sugar concentration demonstrated for glycogen deposition in the present work, is similar to the relationship previously demonstrated for glucose utilization *in vivo*.[1] The presence of this same relationship in both instances is in agreement with our conception that insulin acts by catalyzing the conversion of glucose into some intermediary metabolite which precedes and is necessary for both glycogen formation and sugar utilization.[9] Thus insulin influences the rate at which sugar enters into the metabolic processes of the cell, but the fate of the sugar which has entered depends on other factors, and insulin cannot be regarded as being a *specific* accelerator of either the storage or the oxidation of carbohydrate.

Summary. Glycogen deposition in rat diaphragm *in vitro* varies directly with the concentration of glucose in the medium. Insulin catalyzes this process greatly at low sugar concentrations but very little at high concentrations. The bearing of these results on the mechanism of insulin action is briefly discussed.

12003 P

Effect of Shearing Forces on the Cortex of the Fertilized Egg of *Arbacia punctulata.*

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In previous communications[1, 2] the theory was advanced that, after monaster formation, the cortex of the fertilized egg of *Arbacia punctulata* consists in part of a bound calcium compound. Data were presented showing that this labile combination can be broken down in the living cell by various chemical agents, with the subsequent release

[7] Major, S. B., and Mann, F. C., *Am. J. Physiol.*, 1932, **102**, 409.

[8] Barker, S. B., and Sweet, J. E., *Science*, N. S., 1937, **86**, 270.

[9] Soskin, S., and Levine, R., *Am. J. Physiol.*, 1940, **129**, 782.

[1] Churney, L., *Anat. Rec. Suppl.*, 1938, **72**, 65.

[2] Churney, L., and Moser, F., *Physiol. Zool.*, 1940, **13**, 212.

of free calcium ions. For detecting calcium release, use was made of the observation that the characteristic red pigment "granules" of the egg disintegrate in the presence of calcium or other divalent cations.[3] The purpose of this paper is to describe briefly two observations which indicate that, in addition to the chemical procedures discussed in the previous papers, simple physical means are capable of breaking down the cell cortex and causing calcium release.

When immature eggs, or mature unfertilized eggs, or fertilized eggs that have not reached the monaster stage of development, are slowly crushed by withdrawing the sea water from under the cover slip with filter paper, considerable flattening of the egg can occur without rupture of the cell membrane. The compression can attain such a degree that the cell in many places is only a few micra thick. The red pigment granules remain intact as long as the cell membrane is unbroken. The moment that the cell membrane ruptures, the sea water and protoplasm intermingle and the red chromatophores disintegrate. The results are quite different when fertilized eggs that have reached the monaster stage (10-12 minutes after fertilization) are compressed in a similar manner. The slightest pressure on this egg causes the red pigment granules to "explode" in the region of the cell where the pressure is applied. The cell membrane remains intact and many of the red chromatophores are unbroken. The position of each granule that has disintegrated is marked by the appearance of a clear vacuole. This observation is important as indicating that a reaction has taken place between the granule and a divalent cation. With the application of additional pressure more and more granules disintegrate, until the cell is entirely devoid of them. Yet the degree of flattening of such a cell may be very much less than that of a crushed unfertilized egg in which all the red chromatophores are still intact.

The same general results hold for eggs centrifuged for a short time at low speed; only cells that have reached the monaster stage of development will release pigment to the supernatant fluid.*

In attempting to explain these phenomena the following facts are pertinent. Pigment release is caused by the application of extremely mild centrifugal or shearing forces only after the egg has reached

[3] Heilbrunn, L. V., *Biol. Bull.*, 1934, **66**, 264.

* At certain times of the season the cells are very fragile and centrifuging causes release of pigment from unfertilized eggs. That this is an entirely different phenomenon from the one under consideration is seen in the fact that the supernatant fluid is pink rather than orange. The pink color is typical of cytolyzed eggs in the presence of a high concentration of calcium ions. Such eggs were never used in the experiments.

a certain period of development. At no previous time are these procedures effective; not even when the magnitude of the forces is very much greater. This period of susceptibility coincides with the time of localization of the red pigment granules in a differentiated cortical layer of the cell. Hence it may be deduced that these forces do not act directly on the granules themselves, and it is concluded that the shearing forces act upon the "ground substance" (cortex) in which the granules are imbedded. In doing so they cause the bound calcium component of this cortex to disintegrate. The release of calcium ions is then reflected in the breakdown of the red pigment granules.

12004

Influence of Temperature on the Electrogram and Monophasic Action Potential of the Mammalian Heart.*

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Earlier studies in this laboratory demonstrated that warming or cooling portions of the external surface of the ventricles caused characteristic alterations in the T wave of the electrocardiogram.[1] The basis of these T wave changes was found to be alteration in the duration of one or the other of the two components of the electrocardiogram, *i. e.*, of the dextro- or the levocardiogram. Cold prolonged and heat shortened the duration of these components. The following experiments were undertaken to determine the cause of these alterations in the duration of the dextro- and levocardiograms.

Electrograms and monophasic action potentials were studied in 5 dogs by the method previously described,[2] one lead being taken from the anterior surface of the right ventricle and the other from the anterior surface of the left ventricle. Negativity at the lead on

* This work was aided by a grant from the Fluid Research Funds of the Yale University School of Medicine, and by a Fellowship from the Emanuel Libman Fellowship Fund.

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1 Hoff, H. E., and Nahum, L. H., *Am. J. Physiol.*, 1941, **131**, 700.

2 Hoff, H. E., and Nahum, L. H., PROC. SOC. EXP. BIOL. AND MED., 1940, **45**, 263.