may regulate the metabolism of pyruvate to fatty acids (Denton, Halperin & Randle, 1969).

We acknowledge support from the Medical Research Council, the British Diabetic Association, the British Insulin Manufacturers and the Wellcome Trust.

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Properties of Pyruvate Dehydrogenase from Fat-Cell Mitochondria and its Hormonal Control

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The increased pyruvate dehydrogenase activity in fat-pads exposed to insulin (Coore, Denton, Martin & Randle, 1971) persists in mitochondria prepared from isolated fat-cells by the method of Martin & Denton (1970). Exposure of fat-pads and subsequently cells to insulin increased the pyruvate dehydrogenase/glutamate dehydrogenase activity ratio in extracts of mitochondria prepared from them by $80 \pm 15\%$ (12). Exposure to insulin also increased the rate of conversion of pyruvate into citrate by intact mitochondria incubated with pyruvate + bicarbonate or pyruvate + malate.

Linn, Pettit & Reed (1969) find that pyruvate dehydrogenase from kidney, liver or heart can occur in phosphorylated (inactive) and non-phosphorylated (active) forms, which may be interconverted by an ATP-requiring kinase and an Mg^{2+} -activated phosphatase. Similar transformations of fat-pad pyruvate dehydrogenase can be assumed from the following. In an extract of fat-cell mitochondria prepared in 30 mM-phosphate buffer, pH 7.0, containing 0.5 mM-magnesium chloride, 5 mM-2-mercaptoethanol and 1 μ g of oligomycin/ml the pyruvate dehydrogenase/glutamate dehydrogenase activity ratio was 0.28 throughout incubation for 30 min at 30°C. With addition of 10 mM-magnesium chloride the ratio rose to 0.53 and with addition of 0.1 mM-ATP the ratio fell to 0.04 (within 30 min). The effect of 10 mM-magnesium chloride could be reversed by subsequent dilution and addition of 0.1 mM-ATP, and that of 0.1 mM-ATP by subsequent addition of 10 mM-magnesium chloride. Addition of 5 mM-ATP to intact mitochondria incubated with pyruvate (5 mM), malate (5 mM), carbonyl cyanide *p*-chlorophenylhydrazide (0.5 μ M) and

oligomycin (0.5 μ g/ml) for 5 min was found to decrease the conversion of pyruvate into citrate by 35% and the activity of extracted pyruvate dehydrogenase was halved.

When extracts of mitochondria prepared from adipose tissue with or without insulin were incubated with 10 mM-magnesium chloride before assay the effects of insulin on pyruvate dehydrogenase activity were largely eliminated. This finding, together with the observation that insulin activation of pyruvate dehydrogenase did not alter the K_m for pyruvate (67 μ M), suggests that insulin action may increase the proportion of active (dephospho) pyruvate dehydrogenase. The mechanism of activation by insulin and of reversal by adrenaline are not known. An obvious mechanism is one involving cyclic AMP (adenosine cyclic 3':5'-monophosphate), but no clear inhibitory effects of cyclic AMP on pyruvate dehydrogenase activity in intact mitochondria or mitochondrial extracts have been found.

We acknowledge support from the Medical Research Council, the British Diabetic Association, the British Insulin Manufacturers and the Wellcome Trust.

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An Immuno-electrophoretic Assay of β -Lipoprotein and Albumin in Human Aortic Intima by Direct Electrophoresis from the Tissue Sample into an Antibody-Containing Gel

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In human atherosclerosis much of the lipid accumulated in the artery wall is probably derived from plasma β -lipoprotein. Previously (Smith & Slater, 1970) we measured the immunologically intact lipoprotein by extraction, ultracentrifugation and immuno-plate assay (Fahey & McKelvey, 1965), which required 30–150 mg of tissue. On electrophoresis of minced intima in agarose lipoprotein migrated as a broad and straggling band, but re-mincing and re-running indicated that its removal was mainly complete. This suggested that intimal β -lipoprotein might be measured immunologically by migration into an antibody-containing gel, as suggested by Ressler (1960) for isolated proteins.

Plates (10 cm $\times$ 10 cm) are prepared with 28 ml of