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AN HYPOTHESIS RELATING PHOSPHOLIPID SYNTHESIS IN THE ARTERIAL WALL, DIABETES AND ATHEROSCLEROSIS

It has been a frequent observation that the incidence of atherosclerosis is unusually high among diabetics. Despite this well-known clinical fact, the cause for this relationship is not understood. It has been known for a number of years that the ability to synthesize lipid from carbohydrate is less in the diabetic state. The recent popular belief that the fate of the vasculature is determined by the blood lipids which bathe the vessel wall has made the studies on diabetic liver and adipose tissue of particular interest because of the apparent role they play in regulating certain plasma lipids. Thus, liver slices prepared from alloxan-diabetic rats are almost incapable of forming fatty acids from glucose-C-14,[1] and both glucose oxidation and lipogenesis from glucose are reduced by 80 to 90 per cent in adipose tissue obtained from alloxan-diabetic rats.[2] However, neither studies such as these nor those on blood lipids have provided an adequate explanation for the correlation between atherosclerosis and the diabetic state.

It therefore seems reasonable to look elsewhere for possible clues. One obvious place is the arterial wall itself. It is conceivable that the changes occurring in diabetes can in some manner alter the metabolism of the arterial wall and, in this way, predispose it to the ravages of atherosclerosis. Studies on the metabolism of the arterial wall in normal and diabetic subjects in particular have been limited and, thus, relatively little is known regarding the carbohydrate and lipid metabolism of this tissue. However, the authors would like to present a hypothesis, based on the existing, albeit limited, information, which might relate diabetes and atherosclerosis to alterations in the metabolism of the arterial wall.

Evidence that the arterial wall can synthesize phospholipids is presented in the current issue of this journal[8] (see pages 182 to 188). This investigation corroborates the work of Zilversmit et al.[3,5] and Stein et al.[6,7] who have shown that normal aorta can synthesize phospholipids from various radioactive precursors of the phospholipid molecule such as phosphorus-32, acetate-C-14, and carbon-14 free fatty acids. Additional studies carried out by the authors of this editorial have provided evidence that uniformly labeled glucose-C-14 can also serve as a precursor for arterial wall phospholipids presumably by providing α-glycerophosphate.[9]

The significance of the phospholipid synthetic mech-

anism is unknown, but Zilversmit et al.[10] have shown that the synthesis is accelerated during the development of experimental atherosclerosis. The authors of this editorial have just completed in vitro studies on cholesterol-fed rabbit aortas which also show accelerated incorporation of both radioactive fatty acids and glucose into aorta phospholipids.[9]

Since a number of investigators have suggested that plasma phospholipids may play a role in maintaining the solubility of cholesterol in plasma,[11] it may be similarly possible, as Zilversmit has postulated,[10] that the increased synthesis of aortic phospholipids is an effort to solubilize the cholesterol in the plaque and return it to the plasma. Implied in such a theory is that it might be possible to prevent the accumulation of this relatively insoluble lipid (cholesterol) in the arterial wall, if the synthetic mechanism were capable of accelerating sufficiently.

The enzymatic pathways of phospholipid synthesis of the arterial wall appear to be similar to those proposed for liver.[12,13] The intimate relationship between glucose metabolism and glycerol-containing lipids is well known and is reviewed in the current issue of this journal.[8] Normal glycolysis is required for the generation of α-glycerophosphate, ATP, and fatty acids—all of which are necessary for phospholipid synthesis. Thus, in diabetes it might be predicted that the arterial wall would display a decreased glucose utilization and depressed lipogenesis.

Indeed, Mulcahy and Winegrad[14] have recently shown that glucose uptake and the incorporation of glucose carbon-14 into CO_2, glycogen, and total lipids were markedly decreased in the aortas of alloxan-diabetic rabbits. Urrutia et al.[15] demonstrated that diabetic rat aorta also displayed reduced glucose oxidation, but they were unable to demonstrate, as Foster and Siperstein[16] have done, that fatty acid synthesis from acetate-C-14 was impaired in the aorta of the diabetic. Stein et al.[6] found that when glucose was omitted from the incubation medium, significantly less carbon-14 fatty acid was incorporated into normal aorta phospholipids.

To the authors' knowledge, no investigations have been reported regarding the effects of diabetes on the incorporation of glucose into the phospholipids of the arterial wall. However, it seems reasonable to speculate that the inability of the arterial wall to utilize glucose would deprive it of the various precursors which are necessary for phospholipid synthesis. Thus, the inability of the arteries of the diabetic to accelerate the phospholipid synthetic "defense mechanism," in the face of often elevated blood lipids, might lead to an exaggeration of the atherosclerotic process.

With the use of insulin, it might have been expected that the increased susceptibility of diabetics to develop atherosclerosis would have been diminished but its incidence has remained high. The studies of Mulcahy and Winegrad[14] and Urrutia et al.[15] are particularly interesting in this regard. Both groups of investigators have provided evidence in vitro to show that glucose utilization by the aorta of the diabetic is not sensitive to a direct or immediate effect of insulin. This is to say, that the addition of insulin to incubation flasks containing alloxan-diabetic aortic tissue did not significantly increase the already depressed glucose uptake, CO_2 production or lipogenesis. Work on rabbits recently completed in the authors' laboratory has confirmed these findings for normal aorta.[9] Insulin did not increase glucose oxidation to CO_2 or glucose conversion to phospholipid. This was the case whether uniformly labeled glucose-C-14, glucose-1-C-14, or glucose-6-C-14 were used. Wertheimer and Bentor[17] have presented evidence to the contrary, however, by showing that under somewhat different in vitro conditions, glucose utilization by diabetic rat aorta could be reverted to normal with insulin. Reconciliation of these contradictory studies may be found in Urrutia's work, where it was found that unless great care was taken cleaning adventitial adipose tissue from the vessel, an insulin effect would be observed. Insufficient information is available relative to whether insulin treatment, under in vivo conditions, will correct the metabolic derangements occurring in diabetic blood vessels. Mulcahy and Winegrad, in their studies, tested the effects of insulin in vivo and found that only after forty-two hours of insulin therapy did the depression of glucose utilization by the aorta of the diabetic return to normal.

In conclusion, it must be restated that this hypothesis is unavoidably based on limited information. Most of the investigation has been done on isolated aortic tissue, and this may bear little relationship to the metabolism of intact aorta in the living animal. Further, by necessity, experimental animals have been used, and whether their diabetic condition is equivalent in all respects to that observed in humans is a moot point. Nevertheless, it is anticipated that further studies on the metabolism of the arterial wall will play an important role in our understanding of atherosclerosis and how diabetes may relate to lipid deposition in the arterial wall.

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Who Is Well Nourished?

Before clinical signs of nutritional deficiencies appear, there must be biochemical lesions. The problem that faces biochemists is to develop practical, inexpensive laboratory tests which adequately measure these biochemical lesions. Most medical laboratories do not routinely do many of the laboratory tests used in determining nutritional status. In cases of general malnutrition it is likely that multiple deficiencies occur and that physical, dietary, and anthropometric examinations will provide sufficient information to suggest the proper course of treatment.

Physicians interested in public health nutrition find that biochemical data are of considerable value in determining the nutritional status of groups of people. In recent years the U.S. government, through its Interdepartmental Committee on Nutrition for National Defense, has conducted nutritional surveys in underindustrialized countries all over the world. In general, laboratory tests used in determining nutritional status of populations consist of the measurement in blood or urine of nutrients or products of their metabolism. Such tests may be done with or without load tests of the nutrient being studied. Measurements of enzyme activity or metabolites whose concentration in blood and urine varies with enzyme activity are also done. Hemoglobin, hematocrit, and serum protein levels are also determined. Dr. Gershoff indicated that although these methods are routinely used, many unsolved problems in sampling technics and analytical methods remain.

Many people, because of inborn errors of metabolism, disease conditions, the use of various pharmaceutical agents or an altered physiological state, have nutritional requirements which deviate from what is considered normal. Laboratory investigation of pathologic conditions which fall into these categories is receiving a major share of the attention of nutritional biochemists. Studies in recent years of vitamin B_6 metabolism, for example, have indicated that increased requirements for this vitamin may occur during pregnancy, in some cases of seborrheic dermatitis, in individuals treated with isoniazid, in patients with urinary stones, and in a few convulsive infants with inborn errors of metabolism. In individuals such as these, laboratory tests may be very effective as a diagnostic tool.

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