ABSTRACTS

Ashcroft, S. J. H.; and Randle, P. J. (Univ. of Bristol, Bristol, England): GLUCOSE PHOSPHORYLATING AND GLUCOSE-6-PHOSPHATASE ACTIVITIES OF MOUSE PANCREATIC ISLETS. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5:201-06, 1969.

Verbatim summary. Earlier studies have indicated that the glucose stimulation of insulin release may be mediated by a product of glucose metabolism and that the glucoreceptor may be an enzyme or enzymes controlling the rate of phosphorylation of glucose by the beta cell. We have studied the glucose phosphorylating activities and the glucose-6-phosphatase activity in homogenates of pancreatic islets. The simultaneous operation of a hexokinase inhibited by glucose-6-phosphate and glucose-6-phosphatase inhibited by glucose was found to be capable of constituting a high K_m system for net glucose metabolism. However, a direct test of glucose-6-phosphatase activity in the intact islet suggested that this enzyme is unlikely to be a major factor in the control of glucose phosphorylation.

Results suggesting the presence in islets of a high K_m glucokinase were obtained. The activity of glucokinase was considerably lower than that of hexokinase but may be of prime importance in the response to high glucose.

Bergström, Jonas; Hultman, Eric; Jorfeldt, Lennart; Pernow, Bengt; and Wahren, John (Clin. Central Lab., St. Eriks Hosp. and the Dept. of Clin. Physiol., Karolinska Institutit at Serafimerlasarettet, Stockholm, Sweden): EFFECT OF NICOTINIC ACID ON PHYSICAL WORKING CAPACITY AND ON METABOLISM OF MUSCLE GLYCOGEN IN MAN. J. Appl. Physiol. 26:170-76, February 1969.

Verbatim summary. Fifteen healthy male subjects performed exercise of various durations and intensities on a bicycle ergometer before and after intravenous administration of nicotinic acid (NA) in amounts sufficient to block the release of free fatty acids (FFA) from the adipose tissue. The ability to perform either short-time near maximal work (series A) or prolonged submaximal work (series B) was unchanged before and after administration of NA although all subjects experienced the work after administration of NA as heavier and more fatiguing. The decrease in glycogen content of the quadriceps femoris muscle induced by exercise, as determined in needle biopsy specimens, was significantly larger after administration of NA than in the control studies. The respiratory quotient (RQ) was significantly higher during prolonged exercise after administration of NA compared to the control studies. When the exercise was performed with gradually increased loads up to a near maximal level (series A) the differences in RQ were eliminated at the higher work loads. The arterial concentration of glucose in series A was lower during exercise performed after administration of NA. The results suggest that the reduced delivery of FFA to the muscles after administration of NA is compensated by an increased metabolism of muscle glycogen in such a way that the ability to perform short-term physical work is unaffected.

Briggs, J. H.; Pryor, J. S.; Davison, A. N.; and Fowler, P. B. S. (Charing Cross Hosp., London, England): DIABETES MELLITUS UNMASKED BY ORAL GLYCEROL LOADING. British Diabetic Association Autumn Meeting, 1968; Diabetologia 5:201-06, 1969.

Verbatim summary. A young woman aged twenty with a syndrome of mild diabetes mellitus was found to have normal fasting blood sugars, glucose tolerance and plasma insulin levels. When repeated after priming with 500 mg. glycerol orally, given thirty minutes beforehand, the glucose tolerance test became frankly diabetic, the blood sugar level at two hours being 315 mg./100 ml.

Burch, P. R. J.; and Milunsky, A. (The Gen. Infirmary, Univ. of Leeds, Leeds, England, and Child Development Clin., New England Med. Center Hosps., Boston, Mass.): EARLY-ONSET DIABETES MELLITUS IN THE GENERAL AND DOWN'S SYNDROME POPULATIONS. Lancet 1:554-58, March 15, 1969.

Previous analyses indicate that the prevalence of diabetes in young Down's syndrome patients is much higher than in the general population. In one study diabetes was six times more common during the first decade. This finding permits the postulation that early-onset diabetes may be an autoaggressive (autoimmune) disorder which is initiated in a genetically predisposed person by the occurrence of recessive gene mutations in a single somatic stem cell. This hypothesis fits with the concept that juvenile diabetes is confined to a genetically specific subpopulation characterized by an X-linked factor and two homozygous alleles at two autosomal loci. One allele may be located on chromosome 21. If so, Down's syndrome people predisposed to early-onset diabetes carry it on all three chromosomes. Early-onset diabetes may occur from two random events in a single growth control cell which mutates and propagates a forbidden clone of descendant cells. These cells probably synthesize a mitotic control protein which attacks beta cells immunologically causing the dysfunction characteristic of diabetes mellitus. T.G.S.

Butterfield, W. J. H.; Cox, B. D.; and Whichelow, M. J. (Dept. of Med., Guys Hosp., London, England): THE METABOLIC EFFECTS ON DIABETES OF γ-GUANIDINO-BUTYRAMIDE. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5:201-06, 1969.

Verbatim summary. A new compound, γ guanidinobutyramide (HL 523) with structural similarities to arginine and

phenethylbiguanide has been developed. It is an intermediary in the metabolism of arginine to γ-aminobutyric acid in bacteria but does not occur in man.

HL 523 is nontoxic and hypoglycemic in experimental animals. In man it slightly increases insulin sensitivity in plump nondiabetics, and peripheral glucose uptake to a modest extent in diabetics. However pilot doses of HL 523 for seven to ten days produced no marked hypoglycemia in fifty diabetics.

HL 523 therapy caused a fall of blood urea towards normal in diabetics with elevated levels. A reduction of blood urea has been confirmed in vivo in rats. Studies in man and rats indicate that HL 523 does not affect renal function but that it is acting on the metabolism of the urea cycle amino acids. Studies of glucose and urea cycle amino acids, in the peripheral muscles of normal subjects, show that arginine is released after an overnight fast, and that a single dose of HL 523 increases the release of arginine and lowers the systemic urea level. Guanido compounds may therefore be important in regulating the protein sparing action of dietary carbohydrate and in diabetic complications, particularly renal disease.

Christensen, Halvor N.; and Cullen, Andrea M. (Dept. of Biological Chem., Univ. of Michigan, Ann Arbor, Mich.): BEHAVIOR IN THE RAT OF A TRANSPORT-SPECIFIC, BICYCLIC AMINO ACID. J. Biol. Chem. *244*:1521-26, March 25, 1969.

Verbatim summary. A model amino acid, 2-aminobicyclo (2,2,1) heptane-2-carboxylic acid, highly specific to the Na^+-independent transport agency for neutral amino acids with large apolar side chains, has been shown to be excreted slowly by the intact rat, to be metabolized to a very small extent if at all, and to be accumulated by tissues, especially by pancreas. One of the two isomeric forms of this substance was retained in the rat longer than the other, although both were taken up by tissues, and both presumably were resorbed from the glomerular filtrate. When 5 to 10 mmoles per kg. of body weight were injected intraperitoneally, the urinary excretion of several ordinary amino acids was increased. In addition, the plasma levels of several amino acids were decreased, although that of phenylalanine may have increased. These effects appeared to be at least as severe in adrenalectomized rats, which were easily prostrated by 2-aminobicyclo (2,2,1) heptane-2-carboxylic acid administration. The injections were found to lower the blood glucose, the effect being especially strong in animals first treated with tolbutamide. The effect was more prolonged than that of leucine. This behavior leads us to propose that the stimulation of insulin secretion may be triggered by binding at strategically situated transport receptor sites. The uptake of 2-aminobicyclo (2,2,1) heptane-2-carboxylic acid by the isolated, intact rat diaphragm was, however, not stimulated by insulin.

Christensen, N. J. (Second Clinic of Intern. Med., Kommunehospitalet, Århus University School of Medicine, Århus, Denmark): SPONTANEOUS VARIATIONS IN RESTING BLOOD FLOW, POSTISCHEMIC PEAK FLOW AND VIBRATORY PERCEPTION IN THE FEET OF DIABETICS. Diabetologia *5*:171-78, 1969.

Verbatim summary. The variability of the resting blood flow in the foot was studied by means of venous-occlusion plethysmography in twenty-five diabetics and sixteen nondiabetics. To evaluate the functional state of the vessels themselves, the postischemic peak flow was studied in the same patients. To obtain an estimate of the functional state of the nerves the vibratory perception threshold was determined. The following results were obtained: *1.* The normally occurring spontaneous variations in the resting blood flow in the foot were considerably reduced in the diabetic group of patients, although the mean resting blood flow was identical in the diabetic and the nondiabetic group. The lack of spontaneous variations showed a strong association with the duration of diabetes. *2.* The peak flow was often reduced in long term diabetic patients. This abnormality was associated with the presence of arterial calcifications. *3.* The vibratory perception threshold increased with increasing duration of diabetes as already shown by other authors. *4.* It was demonstrated by statistical analysis that the loss of rhythmic activity in the resting blood flow was due to autonomic neuropathy as well as to a vascular factor.

Corder, C. N.; and Kalkhoff, R. K. (Depts. of Pharmacol. and Med., Marquette Med. Sch., Milwaukee, Wis.): HEPATIC LIPID METABOLISM IN ALLOXAN DIABETIC RATS. J. Lab. Clin. Med. *73*:551-62, 1969.

The effects of the diabetic state and presumably insulin deficiency on hepatic synthesis of triglycerides and cholesterol are little known. Since the liver is the primary source of circulating triglycerides and cholesterol and these lipids are often elevated in clinical diabetes, the effects of alloxan diabetes on hepatic lipid synthesis in the rat were studied. Male rats were made diabetic by intravenous alloxan and studied 3, 6, 11, and 18 days later by analysis of their frozen livers and serum and by in vitro incubation of their unfrozen livers.

Hepatic incorporation of C-14 palmitate and C-14 alpha glycerolphosphate into triglyceride, phospholipid and cholesterol esters was measured and compared to synthetic activities in control rats. The diabetic rats were found to have significantly increased serum triglycerides but no changes in phospholipids or cholesterol at three and eleven days after alloxan. Insulin lowered lipids when given to diabetic animals. Concentration of alpha glycerolphosphate was no different in diabetic than in control livers. Hepatic triglyceride concentrations were lower in diabetic livers. Incorporation of C-14 palmitate into triglyceride was significantly increased at six and eleven days. These data indicate that alloxan diabetes is associated with increased hepatic incorporation of substrate into glyceride. T.G.S.

Davidson, Mayer B.; Lowrie, Edmond G.; and Hampers, Constantine L. (U.S. Army Res. Inst. of Environmental Med., Natick, Mass., Dept. of Med., Peter Bent Brigham Hosp.; and Harvard Med. Sch., Boston, Mass.): LACK OF DIALYZABLE INSULIN ANTAGONIST IN UREMIA. Metabolism *18*:387-94, May 1969.

A technic is described for measurement of insulin antagonism in serum using the rat diaphragm assay in which the activity of insulin added to pre- and post-dialysis sera obtained from eight uremic patients was determined. Intravenous glucose tolerance tests were performed on these patients before and after repeated hemodialysis. In each patient glucose tolerance was improved following dialysis; however, no differences in the activity of insulin added to sera obtained before and after dialysis could be demonstrated by the in vitro assay. These findings suggest that glucose intolerance in uremia is caused by disturbances in metabolism of peripheral tissues rather than the presence of a dialyzable insulin antagonist. C.R.S.

Devlin, J.; and Duggan, M. (Dept. of Med. and Therap., Univ. College, Dublin, and St. Vincent's Hospital, Dublin, Eire): ANTIBODY STUDIES IN PATIENTS ON MIXED BOVINE/PORCINE INSULINS. Diabetologia 5:192-94, 1969.

Verbatim summary. A study of two groups of diabetic sera has been carried out with reference to total insulin binding capacity and preferential binding of beef and pork insulin. Group I (twenty-five patients) received insulin containing predominantly beef insulin with a beef/pork ratio of 3:1 approximately; Group II (thirteen patients) received insulin containing at least 97 per cent pork insulin. In Group I the mean total insulin binding capacity was 15.38 U./liter and in twenty-one beef insulin was preferentially bound with reference to pork insulin. Group II was divided into two subgroups: group IIa, seven patients, the sera bound beef > pork, and the mean insulin binding capacity was 8 U./L. Group IIb, six patients, the sera bound pork > beef or showed no preference, and the mean insulin binding capacity was 3.1 U./L. The mean insulin dose in Group IIa was 34.8 and in Group IIb was 24. The mean insulin binding capacity and mean insulin dose were significantly different ($p < 0.05$) between Group IIa and Group IIb. The possible significance of the results is discussed.

Doeblin, T. D.; Evans, K.; Ingall, G.; Frohman, L. A.; and Bannerman, R. M.: DIABETES IN NORTH AMERICAN INDIANS; THE SENECAS. British Diabetic Association Autumn Meeting, 1968; Diabetologia 5:201-06, 1969.

Verbatim summary. In a systematically selected sample of adult Seneca Indians in western New York, the prevalence of clinical diabetes was found to be 11.6 ± 3.4 per cent among men, and 14.6 ± 3.3 per cent among women. The rest of the sample, not already known to be diabetic, were screened by a plasma glucose determination one hour after a 75 gm. oral glucose load. Twenty per cent of the values obtained were over 200 mg./100 ml. and mean values for various age categories were higher than those of similarly tested general U.S. populations. Values increased with increasing age. Repeat two-hour glucose loading tests with glucose and insulin determinations were also carried out in fifty subjects. Preliminary analysis suggests that diabetes in Seneca Indians is associated with high insulin production.

Henderson, J. R. (Inst. of Psychiatry, Maudsley Hosp., London, England): AN HEPATIC GLUCOSE THRESHOLD. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5: 201-06, 1969.

Verbatim summary. A small but variable concentration of glucose (10-40 mg./100 ml.) has been demonstrated in the bile of most animals—but the phenomenon has never been given much attention. Using anesthetized rabbits and rhesus monkeys, with I.V.C. and biliary catheters, the relationship between plasma and bile glucose has been investigated with a glucose oxidase method. Hyperglycemia was induced by infusing glucose or glucagon.

In the unfasted rabbit, little glucose (< 20 mg./100 ml.) is present in bile, but on increasing blood level above 160—180 mg./100 ml. the bile concentration rises linearly with the plasma concentration, and may reach levels of 200—300 mg./100 ml. In the monkey, glucose appears in bile at lower blood concentrations than in the rabbit, but for technical reasons it is more difficult to show a threshold level. If active glucose transport is blocked with phlorrhizin, then in both species biliary glucose rises to the blood level. It is suggested that under normal circumstances the biliary epithelium actively transports glucose: but this carrier is easily saturated, and glucose then appears in significant quantities in bile.

Hirsch, Jules; and Han, Paul W. (The Rockefeller Univ., New York, N.Y., and the Dept. of Physiol., Sch. of Med., Univ. of Pennsylvania, Philadelphia, Pa.): CELLULARITY OF RAT ADIPOSE TISSUE: EFFECTS OF GROWTH, STARVATION, AND OBESITY. J. Lipid Res. 10:77-82, January 1969.

Verbatim summary. The size, number, and rate of formation of mature adipocytes were studied in the epididymal pads and retroperitoneal adipose depots of the Sprague-Dawley rat. Early growth of these depots was accompanied by progressive enlargement of adipose cells as well as by increases in number. Beyond the fifteenth week of life, the depot grew exclusively by the process of cellular enlargement, wtih no further change in cell number. Severe starvation during the sixth week of life followed by normal feeding had no lasting effect on cell size or cell number; prolonged semistarvation beginning in the fifteenth week greatly reduced cell size while cell number was unaffected. Likewise, extreme increases in depot size produced by hypothalamic lesions did not change cell number, but only cell size. The concept of a fixed number of mature adipocytes in the adult organism may be of central importance in caloric and metabolic equilibrium.

Jeffcoate, S. L.; and Moody, A. J. (NOVO Research Institute, Copenhagen, Denmark): THE DISPOSAL OF ORALLY ADMINISTERED C-14-GLUCOSE IN THE NORMAL RAT. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5: 201-06, 1969.

Verbatim summary. 6-C-14-glucose (specific activity 6.0 μC./gm.) was administered by esophageal tube to fasted normal rats; the load was 1.5 gm./kg. Rats were killed, in groups, at 0, 10, 20, 30, 60, 120 and 180 minutes after administration of the load, and the blood and liver sampled. Serum insulin and serum glucose peaked at twenty minutes. The radioactive serum glucose measured by the radioactivity in the dimedone derivatives of 6-C of glucose peaked at sixty minutes. At this time 92 per cent of the serum glucose originated from the load. Liver glycogen started to rise after ten to twenty minutes, reaching a maximum at 120 minutes. The incorporation of C-14-glucose into liver glycogen paralleled the changes in total glycogen.

These data are analyzed. It is suggested that during the first hour following an oral glucose load there is a reduction in the hepatic release of glucose accompanied by an increase in hepatic glycogen synthesis. During the second hour there is a further marked increase in hepatic glycogen. Between 120 and 180 minutes there is a decrease in hepatic glycogen, and hepatic glucose release increases. The results emphasize the important role of the liver in the disposal of an oral glucose load.

Mancini, A. M.; Zampa, G. A.; Geminiani, G. D.; and Vecchi, A. (Instit. of Morbid Anat. of the Univ. of Sassari and Endocrine and Metabolic Unit of Bentivoglio Hosp., Bologna, Italy): EXPERIMENTAL NODULAR "DIABETIC-LIKE" GLOMERULOSCLEROSIS IN GUINEA PIGS FOLLOWING LONG-ACTING, HETEROLOGOUS INSULIN IMMUNIZATION. Diabetologia 5: 155-66, 1969.

Verbatim summary. Renal lesions similar to Kimmelstiel-Wilson nephropathy have been found in guinea pigs immunized with long-acting heterologous insulin. The animals were treated for periods ranging from three to five months, with monthly subcutaneous injections of highly purified bovine insulin in a mixture of lanolin and paraffin oil. Kidney sections have been examined by means of light microscopy after standard and histochemical stainings, histoimmunological technics and electron microscopy. The following glomerular lesions have been detected: *1.* PAS-positive hyaline nodules (55.5 per cent); *2.* Thickening of the basal membrane (100 per cent); *3.* aneurysmatic dilatation of the capillaries (88.8 per cent); *4.* diffuse glomerulosclerosis (61.1 per cent); *5.* increased number of mesangial cells (38.8 per cent); *6.* fibrinoid caps (44.4 per cent); *7.* capsular adhesions (33.3 per cent).

Histochemical findings showed that there are differential characteristics between fibrinoid caps (exudative lesions) and hyaline nodular alterations: the former were rapidly digested by trypsin, whereas the nodules resisted the tryptic treatment for six hours. Histoimmunological staining of the lyophilized kidney sections with fluorescein-isothiocyanate-labeled anti-bovine insulin serum showed marked fluorescence of both the fibrinoid caps and hyaline nodules of the glomeruli, and also a less intense fluorescence of the basement membrane and of the intercapillary stroma.

Electron microscopy of the glomeruli demonstrated that the basal membrane was always irregularly thickened showing a patchy fibrillary structure, and that there were nodular areas and bands of "basal-membrane-like" material. Besides, dense osmiophilic deposits were noted in the mesangium. It is concluded that insulin acting as an antigen brings about a nodular Kimmelstiel-Wilson-like nephropathy in experimental animals with normal or low blood sugar and lipids, through immunological mechanisms.

Summers, R. O. C.; Soler, N. G.; and FitzGerald, M. G. (Diabetic Clinic, The General Hosp., Birmingham, England): RETINOPATHY IN YOUNG DIABETICS. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5:201-06, 1969.

Verbatim summary. The incidence of retinopathy present at diagnosis varies in different reports from 4 per cent to 15 per cent. From 1960 to 1967, 5,157 new diabetics were seen in whom retinopathy was present in 389 (7.5 per cent). Retinopathy is acknowledged to be a late complication of diabetes and, when present at diagnosis, probably results from asymptomatic longstanding mild diabetes. There are ten patients under forty years of age in this group with retinopathy at diagnosis, and their clinical features are compared with those of their older counterparts. Only one patient presented with a classical symptom of diabetes-pruritus vulvae. Two were diagnosed following crops of boils, one with a neuropathic ulcer of the foot and two were referred following ophthalmic examination. Four were found quite incidentally on urine testing. Two years was the maximum duration of evidence of diabetes, in one case only. No patients presented with acute diabetes. Of the ten patients, five had at least one other abnormality (neuropathy, proteinuria, hypertension, claudication).

The evidence is in favor of these patients having had longstanding mild asymptomatic diabetes. Their subsequent clinical course does not suggest a type of diabetes with accelerated onset of complications. This was often related to uncontrolled diabetes, but sometimes occurred in association with normal blood sugars when patients were vomiting or starving. Other patients with severely uncontrolled diabetes had normal blood ketone levels. More extensive application of this method should result in a greater appreciation of the significance of hyperketonemia and ketonuria.

Turner, R. C.; Oakley, N. W.; Nabarro, J. D. N.; Coltart, T.; and Beard, R. W. (Middlesex and Queen Charlotte's Hospitals, London, England): BLOOD GLUCOSE AND INSULIN RELATIONSHIPS IN THE HUMAN MOTHER AND FETUS. British Diabetic Association Autumn Meeting, 1968. Diabetologia 5: 201-06, 1969.

Verbatim summary. Intravenous glucose tolerance tests were performed on eight pregnant patients at the time of surgical induction of labor. Fetal scalp capillary blood, obtained by Saling's method, and maternal venous blood were collected over a ninety-minute period. The plasma was assayed for glucose and immunoreactive insulin.

The plasma glucose in the fasting state was the same in the fetus as in the mother. The fetal level showed a marked increase by five minutes after the injection and was maximal at ten minutes. The level then approached the maternal level, and in the latter part of the test the fetal capillary glucose was on the average 7 mg./100 ml. less than the maternal venous glucose. This suggests the transplacental gradient may be less than previously reported.

The initial fetal plasma insulin levels varied from 5-30 μU./ml. In four fetuses there was a rise in the plasma insulin at five minutes, and in one fetus a delayed rise at one hour after the injection. There was no correlation of the fetal insulin with the maternal insulin levels. It is unlikely that maternal insulin crosses the placenta and it is thought the rise in fetal insulin is a response of the fetal pancreas to glucose.

Whitty, Albert J.; Shima, Kenji; Trubow, Marshall; and Foà, Piero P. (Div. of Res., Sinai Hosp. of Detroit, Detroit, Mich.): EFFECT OF GLUCAGON AND OF INSULIN ON SERUM FREE FATTY ACIDS IN NORMAL AND DEPANCREATIZED DOGS. Proc. Soc. Exp. Biol. Med. 130:55-61, January 1969.

Verbatim summary. Intravenous injections of glucagon were followed by a triphasic response in serum FFA concentration: an immediate rise, probably reflecting the lipolytic effect of the hormone, a secondary depression, probably caused by hyperglycemia and by exogenous and/or endogenous insulin, and a final rise, which may have been the result of continued glucagon-induced lipolysis. It may be concluded that although glucagon depresses serum FFA concentration in vivo by stimulating insulin and glucose release, under suitable circumstances, its lipolytic effect can be demonstrated by a rise in serum FFA. A dose of insulin, as small as 0.5 mU./kg., which caused little change in serum glucose, was found to be effective in reducing serum FFA concentration. This amount of insulin may be present in commonly used doses of crystalline glucagon and may contribute significantly to their biologic effects. The prompt and potent insulogenic action of glucagon in vivo was confirmed. Three to four days after total pancreatectomy, serum IRI had decreased but had not disappeared. In these animals, the IRI response to glucagon could not be demonstrated.