sen, M., and Svejgaard, A.: HL-A antigens and diabetes mellitus. Lancet *2*:864-66, 1974.

[140]Creutzfeldt, W., Kobberling, J., and Neel, J. V., Editors: The Genetics of Diabetes Mellitus. New York, Springer Verlag, 1976.

[141]Thomsen, M., Platz, P., Anderson, O. O., Christy, M., Lyngsøe, J., Nerup, J., Rasmussen, K., Ryder, L. P., Nielsen, L. S., and Svejgaard, A.: MLC typing in juvenile diabetes mellitus and idiopathic Addison's disease. Transplant. Rev. *22*:125-47, 1975.

[142]Ludwig, H., Schernthaner, G., and Mayr, W. R.: Is HLA-B7 a marker associated with a protective gene in juvenile-onset diabetes mellitus? N. Engl. J. Med. *294*:1066, 1976.

[143]Barbosa, J., Noreen, H., Emme, L., Goetz, F., Simmons, R., deLeiva, A., Najarian, J., and Yunis, E. J.: Histocompatibility (HLA) antigens and diabetic microangiopathy. Tissue Antigens *7*:233-37, 1976.

A[144]Rubinstein, P., Suciu-Foca, N., and Nicholson, J. F.: Genetics of juvenile diabetes mellitus: A recessive gene closely linked to HLA D and with 50 per cent penetrance. N. Engl. J. Med. *297*:1036-40, 1977.

[145]Barbosa, J., King, R., Noreen, H., and Yunis, E. J.: The histocompatibility system in juvenile, insulin-dependent diabetics multiplex kindreds. J. Clin. Invest. *60*:989-98, 1977.

[146]deVries, R. R. P., Kreeftenberg, H. G., Loggen, H. G., and van Rood, J. J.: In vitro immune responsiveness to vaccinia virus and HLA. N. Engl. J. Med. *297*:692-97, 1977.

A[147]Christau, B., Kromann, H., Andersen, O. O., Christy, M., Buschard, K., Arnung, K., Kristensen, I. H., Peitersen, B., Steinrud, J., and Nerup, J.: Incidence, seasonal and geographical patterns of juvenile-onset insulin-dependent diabetes mellitus in Denmark. Diabetologia *13*:281-84, 1977.

A[148]Cudworth, A. G., Gamble, D. R., White, G. B. B., Lendrum, R., Woodrow, J. C., and Bloom, A.: Etiology of juvenile-onset diabetes: A prospective study. Lancet *1*:385-88, 1977.

[149]Kibrick, S., and Benirschke, K.: Severe generalized disease (encephalo-hepatomyocarditis) occurring in the newborn period and due to infection with Coxsackie virus, group B. Pediatrics *22*:857-75, 1958.

[150]Vizso, A. D., and Sanders, F. K.: Alteration of the pathogenicity of some group B Coxsackie viruses under different conditions of passage. I. Virus type 4. Acta Virol. (Praho) *8*:38-45, 1964.

[151]Munterfering, H.: Zur Pathologie des Diabetes Mellitus der weissen Maus bei der EMC-Virusinfektion: Histologishe, elektron-mikroskopische und quantitativ morphologische Befunde an den Langerhanssacken Inseln. Virchows Arch. A *356*:207-34, 1972.

[152]Wellmann, K. F., Amsterdam, D., Brancato, P., and Volk, P. W.: Fine structure of pancreatic islets of mice infected with the M variant of the encephalomyocarditis virus. Diabetologia *8*:349-57, 1972.

[153]Platt, H.: Observations on the pathology of experimental foot-and-mouth disease in the adult guinea pig. J. Pathol. Bacteriol. *76*:119-31, 1958.

[154]Platt, H.: A study of the pathological changes produced in young mice by the virus of foot-and-mouth disease. J. Pathol. Bacteriol. *72*:299-312, 1956.

[155]Robertson, J. S.: The pancreatic lesion in adult mice infected with a strain of pleurodynia virus. I. Electron microscopical observations. Aust. J. Exp. Biol. Med. Sci. *32*:393-409, 1954.

ABSTRACTS

(All are verbatim summaries, except those from Nature*)*

Prince, G. A.; Jenson, A. B.; Billups, L. C.; and Notkins, A. L. (Lab. of Oral Med., Natl. Inst. of Dental Res., NIH, Bethesda, Md.): INFECTION OF HUMAN PANCREATIC BETA CELL CULTURES WITH MUMPS VIRUS. Nature (London) *271*:158-61, 1978.

Human pancreatic cultures from seven autopsy subjects (both sexes) ranging in age from 3 days to 46 years were examined for their susceptibility to mumps virus. The proportion of beta cells in these cultures, determined by staining with rhodamine-labelled anti-insulin antibody, ranged from 1-5%.

The cultures were infected with the ABC strain of mumps virus and at various times thereafter stained by the double-label antibody technique using rhodamine-labelled anti-insulin antibody and fluorescein-labelled anti-mumps antibody.

60-90% of the beta cells in these infected cultures contained mumps antigen in the cytoplasm. Similarly, 60-95% of non-insulin containing cells in the same cultures also contained mumps antigens. The effect of mumps virus replication on pancreatic cell survival was also examined by following these infected cultures daily for 6 days. Virus titers increased by 72 hours after infection, and then declined. At 6 days after infection, the ratio of the number of beta cells in infected cultures to the number of beta cells in uninfected cultures decreased from 1 to 0.1. The highly lytic nature of the mumps infection points to beta cell death as the most likely explanation for this decrease rather than virus-induced degranulation. Thus, by use of the double-label antibody technique, it is shown that human beta cells in vitro are susceptible to infection by mumps virus.

Menser, M. A.; Forrest, J. M.; and Bransby, R. D. (Children's Med. Res. Found., Royal Alexandra Hosp. for Children, Camperdown, New South Wales, Australia): RUBELLA INFECTION AND DIABETES MELLITUS. Lancet *1*:57-60, 1978.

The incidence of diabetes mellitus was increased in patients with congenital rubella. Experimental congenital rubella infection in rabbits caused histological changes in the β-cells of the pancreatic islets similar to those found in mice made diabetic by the M variant of the encephalomyocarditis virus. It is concluded that the diabetes seen in congenital rubella is due to viral infection of the pancreatic islet cells.

Rossini, A. A.; Like, A. A.; Chick, W. L.; Appel, M. C.; and Cahill, G. F., Jr. (Joslin Res. Lab., Harvard Med. Sch., and Peter Bent Brigham Hosp., Boston, and Dept. of Pathol., Univ. of Mass. Med. Sch., Worcester, MA): STUDIES OF STREPTOZOTOCIN-INDUCED INSULITIS AND DIABETES. Proc. Natl. Acad. Sci. USA *74*:2485-89, 1977.

Multiple small injections of streptozotocin produce a delayed, progressive increase in plasma glucose in mice within 5-6 days after the injections, in association with pronounced insulitis and induction of type C viruses within beta cells. Multiple subdiabetogenic doses of streptozotocin in rats and multiple injections of another beta cell toxin, alloxan, in mice did not induce insulitis although hypoglycemia followed the injection of larger quantities of both agents. In mice, the prior injection of 3-0-methyl-D-glucose (3-OMG) or nicotinamide attenuated the diabetic syndrome produced by streptozotocin; however, 3-OMG was more protective. Rabbit antimouse lymphocyte serum, alone,

provided partial protection but, when given together with either 3-OMG or nicotinamide, effectively prevented the streptozotocin-induced diabetic syndrome. Cessation of these preventive treatments was followed by the appearance of insulitis and diabetes. These findings suggest that multiple injections of streptozotocin induce, in susceptible hosts, the triad of direct beta cell cytotoxicity, virus induction within beta cells, and cell-mediated autoimmune reaction. These factors, acting separately or in concert, appear to induce a destructive insulitis and severe diabetes. The relative importance of each component and the factors governing host susceptibility remain to be clarified.

Yoon, J. W.; Lesniak, M. A.; Fussganger, R.; and Notkins, A. B. (Lab. of Oral Med., Natl. Inst. of Dental Res., NIH, Bethesda, Md.): GENETIC DIFFERENCES IN SUSCEPTIBILITY OF PANCREATIC B CELLS TO VIRUS-INDUCED DIABETES MELLITUS. Nature (London) *264*:178-80, 1976.

The development of encephalomyocarditis (EMC) virus-induced diabetes is genetically determined, with susceptibility being inherited as an autosomal recessive trait. Mice susceptible (SWR/J) and resistant (C57BL/6J) to the development of EMC-induced diabetes, as well as the F1 and F2 offspring were infected with the M variant of EMC virus, and the capacity of EMC virus to replicate in beta cells from each strain as well as the effect of the viral infection on blood insulin and glucose levels were investigated.

Within 48 hours after infection, almost two-thirds of the SWR/J mice had insulin and glucose values outside the normal range of the control group. At 14 days, many of these mice became hypoinsulinemic and hyperglycemic. In contrast to the SWR/J mice, the glucose and insulin values of the C57BL/6J and F1 hybrids generally fell within the normal range, and F2 mice became segregated into two groups—one with glucose and insulin levels within the normal range and the other showing severe hypoinsulinemia and hyperglycemia.

The viral titer in beta cells isolated from each mouse showed the greatest difference between SWR/J and C57BL/6J at 48 hours after infection and was higher in males than in females. The mean viral titer in the beta cells of F1 hybrids was not significantly different from that of the C57BL/6J mice, but that in F2 hybrids was significantly different from F1 hybrids and C57BL/6J mice. With some exception, mice with the highest viral titer in the pancreatic beta cells had the highest circulating insulin and lowest glucose levels. In contrast, the resistant C57BL/6J and F1 hybrid mice had viral titers below 10^5 PFU and their glucose and insulin levels fell within the normal range; F2 hybrid mice segregated into both types. These results suggest that EMC-induced hypoinsulinemia and hyperglycemia are secondary to genetically determined differences in the permissiveness of beta cells to support viral replication.

Yoon, J.; Onodera, T.; and Notkins, A. L. (Lab. of Oral Med., Natl. Inst. of Dental Res., NIH, Bethesda, Md.): VIRUS-INDUCED DIABETES MELLITUS. VIII. PASSAGE OF ENCEPHALOMOCARDITIS VIRUS AND SEVERITY OF DIABETES IN SUSCEPTIBLE AND RESISTANT STRAINS OF MICE. J. Gen. Virol. *37*:225-32, 1977.

The diabetogenic capacity of the M-variant of encephalomyocarditis (EMC) virus was markedly diminished after passage in mouse kidney cell cultures. One passage in mice fully restored this capacity. Virus harvested after five passages in either susceptible (SWR/J) or resistant (C57BL/6J) strains of mice was capable of producing diabetes in susceptible SWR/J mice but not in resistant C57BL/6J mice. Resistance was not overcome by inoculating mice with high concentrations of virus. Immunofluorescence studies showed that islets from strains of mice (i.e. CBA,AKR,C57BL/6J, A/J) that did not develop diabetes after infection with EMC virus, nonetheless, contained virus antigens. The percentage of cells in the islets containing virus antigens varied from 3.6% in CBA to 13.5% in A/J. In contrast, 38% of the islet cells in susceptible SWR/J mice contained virus antigens. It is concluded that both the genetic background of the host and the passage history of the virus influence the development of diabetes.

Chairez, R.; Yoon, J-W.; and Notkins, A. L. (Lab. of Oral Med., Natl. Inst. of Dental Res., NIH, Bethesda, Md.): VIRUS-INDUCED DIABETES MELLITUS. X. ATTACHMENT OF ENCEPHALOMYOCARDITIS VIRUS AND PERMISSIVENESS OF CULTURED PANCREATIC BETA CELLS TO INFECTION. Virology. In press, 1978.

Monolayers of pancreatic beta cells from strains of mice susceptible (SJL/J) and resistant (C57BL/6J) to the development of virus-induced diabetes mellitus were inoculated with the M-variant of encephalomyocarditis (EMC) virus. Immunofluorescence showed that viral antigens appeared in up to 10 times more beta cells from susceptible SJL/J mice than resistant C57BL/6J mice. Infectious center assays revealed that 10-30 times more SJL/J beta cells contained infectious virus than C57BL/6J beta cells. Viral attachment experiments showed no difference in the binding of EMC virus when embryonic fibroblasts, pancreatic fibroblasts, and kidney cells from SJL/J and C57BL/6J mice were compared. However, at least two times more virus attached to pancreatic beta cells from susceptible than resistant strains of mice. Our data suggest that genetically determined differences in viral receptors on the surface of beta cells may be one of the factors controlling susceptibility to EMC-induced diabetes mellitus.

Jansen, F. K.; Münterfering, H.; and Schmidt, W. A. K. (Diabetes Res. Inst., Pathol. Inst., and Inst. for Med. Microbiol. and Virol. of the Univ. of Dusseldorf, Germany): VIRUS INDUCED DIABETES AND THE IMMUNE SYSTEM. I. SUGGESTION THAT APPEARANCE OF DIABETES DEPENDS ON IMMUNE REACTIONS. Diabetologia *13*:545-49, 1977.

The participation of immune reactions in the EMC virus-induced diabetes of the mouse was studied by immunosuppression with 500 R sublethal X-irradiation or 120 mg/kg Asta 5122, a cyclophosphamide derivative. Average glucose levels after X-irradiation and infection remained normal, while virus-infected, otherwise untreated mice, had significantly higher mean glucose levels, indicating that immune reactions are necessary for the development of virus induced diabetes. Immune suppression by the cyclophosphamide derivative led, in contrast, to a significantly increased mean glucose level and increased insulitis in comparison with the controls only infected. This indicates an important role of the cellular immune reaction, insulitis, in the destruction of the islets.

Rayfield, E. J.; Gorelkin, L.; Curnow, R. T.; and Jahrling, P. B. (U. S. Army Med. Res. Inst. of Infect. Dis., Frederick, Md., and Mt. Sinai Sch. of Med., NY, NY): VIRUS-INDUCED PANCREATIC

DISEASE BY VENEZUELAN ENCEPHALITIS VIRUS: ALTERATIONS IN GLUCOSE TOLERANCE AND INSULIN RELEASE. Diabetes *25*:623-31, 1976.

Viral infections have been implicated in the induction of diabetes mellitus in man and laboratory animals. Since virus-specific immunofluorescence (FA) is detectable in hamster pancreas during the acute phase of Venezuelan encephalitis (VE), experiments were designed to correlate pathologic and virologic events with metabolic studies in VE-infected hamsters. Golden Syrian hamsters were inoculated s.c. in groups of four to 12 with 100,000 plaque-forming units (PFU) of the vaccine strain (TC-83) of VE or 1,000 PFU of the virulent Trinidad strain of VE. Ultrastructurally, during Trinidad infection, mature virions were associated with the cell surfaces and within pancreatic beta cells in contrast to absence of virus-related changes in TC-83-infected hamsters. Virus-specific-FA was noted in islet cells and acinar cells of Trinidad-infected hamsters. VE growth curves demonstrated viral replication in pancreas with both strains. Although ultrastructural and FA changes were much more prominent in Trinidad-infected hamsters in contrast to TC-83-infected hamsters during the first few days of illness, the rapid lethality of the Trinidad-infected group necessitated performing all metabolic studies in TC-83-strain-infected hamsters. Accordingly, for the metabolic studies, glucose tolerance tests (GTT) using 2 gm. or 5 gm./kg. glucose i.p. were performed in groups of hamsters acutely infected two days earlier with the TC-83 vaccine strain and in 24-day and 90-day convalescent hamsters after TC-83 vaccine strain. Samples were obtained for glucose and immunoreactive insulin (IRI) determinations. Glucose intolerance occurred in hamsters in each of the infected groups given 5 gm./kg. glucose except for the 90-day convalescent TC-83 group. Severely decreased IRI responses occurred in the 24-day and 90-day convalescent TC-83 hamsters following both 2- and 5-gm./kg. glucose. Pancreatic IRI content in 24-day convalescent TC-83 hamsters was within normal limits, suggesting a defect in IRI release from the beta cells at this stage of convalescence.

Lendrum, R.; Walker, G.; Cudworth, A. G.; Theophanides, C.; Pyke, D. A.; Bloom, A.; and Gamble, D. R. (Dept. of Gastroenterol., St. Mary's Hosp., London, Dept. of Med., King's Coll. Hosp., London, Whittington Hosp., London, and Public Health Lab., West Park Hosp., Epsom, Surrey, England): ISLET-CELL ANTIBODIES IN DIABETES MELLITUS. Lancet *2*:1273-76, 1976.

Islet-cell antibodies (I.C.A.) were found in 38% (319/829) of insulin-dependent diabetic patients, in 5% (6/112) of insulin-independent diabetics, and in 1.7% (3/177) of non-diabetic subjects. In the insulin-dependent group I.C.A. were found in 85% of patients immediately after the onset of symptoms and they became less common as the duration of disease increased. I.C.A. were equally common in both sexes and the decline in their prevalence was independent of age. The antibodies were directed against cytoplasmic components of islet cells but not against insulin itself. The appearance of I.C.A. probably follows cell damage occurring before the onset of symptoms. By contrast, thyroid and gastric autoantibodies were more common in older patients and females. There was no correlation between the presence of these antibodies and I.C.A. in patients with either diabetes of recent onset or longstanding disease.

Rubinstein, P.; Suciu-Foca, N.; and Nicholson, J. F. (Lindsley F. Kimball Res. Inst. of N. Y. Blood Ctr. and Coll. of Physicians and Surgeons of Columbia Univ., New York, NY): GENETICS OF JUVENILE DIABETES MELLITUS: A RECESSIVE GENE CLOSELY LINKED TO HLA D AND WITH 50 PER CENT PENETRANCE. N. Engl. J. Med. *297*:1036-40, 1977.

We investigated the genetic predisposition to juvenile diabetes in the families of 31 index cases in relation to the inheritance of the HLA system. The diabetes-predisposing gene was found to be recessive because the diabetic sibs in index cases shared both their HLA genes with a significantly increased frequency. Penetrance was estimated at 50 per cent because half the HLA-identical sibs in index cases were diabetic. These conclusions fit with published observations that the risk to sibs of patients is about 10 per cent when both parents are normal.

In three informative cases of recombination with HLA the predisposing gene traveled with the HLA segment of the recombinant haplotype. We prepared tables for the computation of risks to relatives based on the hypothesis of recessivity, HLA linkage and 50 per cent penetrance.

Christau, B.; Kromann, H.; Andersen, O. O.; Christy, M.; Buschard, K.; Arnung, K.; Kristensen, I. H.; Peitersen, B.; Steinrud, J.; and Nerup, J. (Steno Mem. Hosp., Gentofte, Med. Dept. E., Frederiksberg Hosp., Copenhagen, Med. Dept. Hvidovre Hosp., Med. Dept. F, Herlev Hosp., Herlev, Dept. of Med. and Pediatr., Frederiksborg County Hosp., Hillerod and Children's Hosp., Fuglebakken, Copenhagen, Denmark): INCIDENCE, SEASONAL AND GEOGRAPHICAL PATTERNS OF JUVENILE-ONSET INSULIN-DEPENDENT DIABETES MELLITUS IN DENMARK. Diabetologia *13*:281-84, 1977.

The incidence, sex, seasonal and geographical patterns of juvenile-onset insulin-dependent diabetes mellitus (j.i.d.m.) were studied retrospectively on one third of the Danish population 1970-1974. The j.i.d.m. incidence remained fairly constant during the study period, the average being 13.2 per 100,000 per year. The total number of boys exceeded the number of girls by 27 per cent. A marked peak of incidence was found at 12-14 years, earlier for females than for males. A seasonal variation in onset (diagnosis) of j.i.d.m. was observed with the lowest number of new cases in May-July. The j.i.d.m. incidence seemed to show socio-economic differences, being highest in those parts of the survey area with lower status.

Cudworth, A. G.; Gamble, D. R.; White, G. B. B.; Lendrum, R.; Woodrow, J. C.; and Bloom, A. (Dept. of Med., Univ. of Liverpool, England): ETIOLOGY OF JUVENILE-ONSET DIABETES: A PROSPECTIVE STUDY. Lancet *1*:385-88, 1977.

110 people in whom insulin-dependent diabetes developed when they were less than 30 years old were studied as soon as possible after diagnosis. There was evidence for clustering of cases with BW15-positive phenotypes during the winter peak (1976) but not during the autumn peak (1975). Subjects who were BW15-positive, and in particular those who were both B8 and BW15 positive, had higher neutralising antibody titres to Coxsackie virus types B1-B4. 58% of cases had islet-cell antibodies (I.C.A.), but the presence of I.C.A. was not correlated with HLA phenotypes or viral antibody titres. In 41 subjects (37%), who gave a definite history of antecedent illness, evidence indicated that this was a precipitating infection and not the initiating event producing islet-cell damage. Nearly half the subjects had had diabetic symptoms for more than 4 weeks before diagnosis.