Metabolic Alterations in Adulthood After Intrauterine Development in Mothers With Mild Diabetes

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We studied the long-term effects of maternal diabetes mellitus on the offspring of experimentally induced diabetic Wistar rats. When stressed by an intravenous glucose load, the adult female offspring had impaired glucose tolerance and developed gestational diabetes mellitus when pregnant. Our results show that even mild diabetes mellitus induces an abnormal intrauterine milieu that causes morphological and functional changes in fetal development with consequences for later life. *Diabetes* 40 (Suppl. 2): 106–108, 1991

Maternal diabetes mellitus involves an abnormal intrauterine milieu for the developing fetus. The disturbed development of the fetal endocrine pancreas and the adaptation of fetal metabolism to the abundant fuel supply might have persistent consequences in these offspring in later life. In this respect, we proposed earlier that gestational diabetes mellitus is an acquired condition and that the diabetic intrauterine milieu in itself may be responsible for the transmission of diabetes mellitus from one generation to another, without interference of a genetic factor.

EXPERIMENTAL MODEL

We studied two subsequent generations of Wistar rats born to experimentally induced diabetic pregnant first-generation rats. A single 30-mg/kg body wt dose of streptozocin was injected on the day of mating in the first-generation animals. The result was a slight increase in the blood glucose, which became significant by the end of gestation. We examined the fetuses of these first-generation mothers at day 20 and also the female adult offspring 100 days after birth in nonpregnant and pregnant states. The offspring of the second-generation mothers consisted of the third generation and were examined as fetuses and adult offspring.

As previously shown, fetuses of the second generation were stressed by mild hyperglycemia, resulting in hyperplasia of the islets of Langerhans, hyperactivity of the insulin-producing β-cells, and hyperinsulinism (1). The total amino acid concentration was reduced in fetal plasma, whereas it was in the normal range in their mildly diabetic mothers. This fact suggests an increased amino acid turnover; furthermore, fetuses of mildly diabetic mothers were macrosomic.

After birth, the abundant maternal food supply is missing. β-Cell mass and insulin secretion are reduced. Twenty days after birth, at the time of weaning, hyperglycemia was present, and the pups remained heavier than the controls. Some of the pups were allowed to grow to adulthood and were examined at 100 days of age. These adult female offspring of the second generation showed normal basal glycemia levels and a normal morphological aspect of the endocrine pancreas. However, when stressed by an intravenous glucose load, impaired glucose tolerance was present in these animals, and when pregnant, they developed mild gestational diabetes mellitus (2,3).

From our early experimental studies, it can be concluded that even mild diabetes in the mother causes a diabetogenic tendency in the offspring over subsequent generations. The origin of these alterations in the second and third generations must be attributed to the abnormal diabetic intrauterine environment during the first-generation pregnancy. However, at that time, we could not exclude streptozocin's effect on the genetic material of the offspring. To answer this question, we programmed four groups of third-generation offspring: *1*) fetuses born to nondiabetic parents and grandparents, *2*) fetuses born to a nondiabetic mother and a father who was born to a streptozocin-induced diabetic mother, *3*) fetuses born to a mother who was born to a diabetic mother and a nondiabetic second-generation fa-

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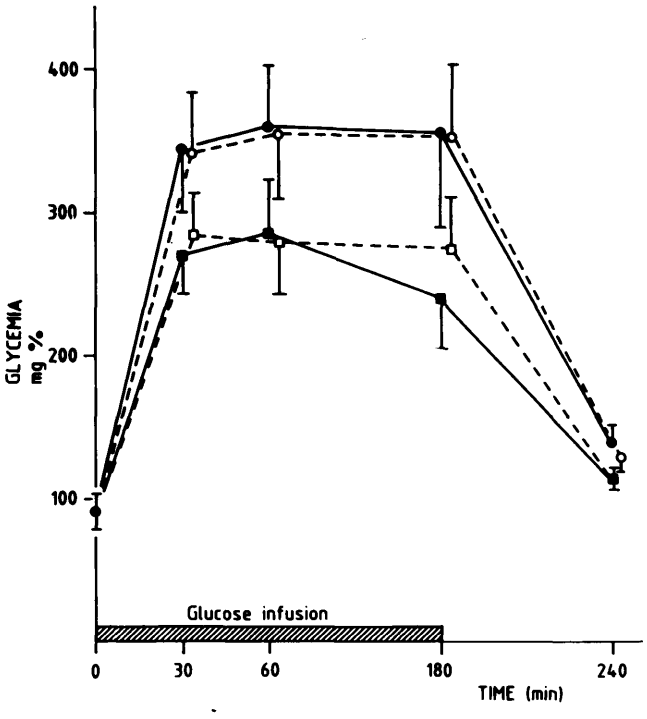

FIG. 1. Mean ± SD plasma glucose concentrations during in vivo glucose infusion (30%, 1.5 ml/h) in 3rd-generation adult offspring with control mother and father (■), control mother and father born to mildly diabetic mother (□), mother born to mildly diabetic mother and control father (●), and mother and father born to mildly diabetic mother (○).

ther, and *4*) fetuses with a second-generation father and mother born to a first-generation diabetic mother.

If streptozocin injection in the first generation had altered the genetic material of the offspring in the second-generation rats, the diabetogenic effect in the second generation should have been expressed in the female and the male lines. However, the typical morphological features of the fetal endocrine pancreas are only present when the second-generation mother is born to a first-generation diabetic mother. Indeed, an increased amount of endocrine tissue and an augmented degranulation of the β-cells of third-generation fetuses are only present in the female line (4).

Moreover, when these four groups became adults and were submitted to a glucose load, an abnormal test was present only in third-generation adult offspring with a mother born to a diabetic mother, regardless of the origin of the father (5; Fig. 1). We have concluded from these data that the diabetic intrauterine milieu is responsible for the changes in subsequent generations. In our experimental model, a genetic factor must not be taken into account.

Experimental data in the rat have been confirmed in human studies. Indeed, epidemiological studies agree that impaired glucose tolerance in the offspring can be a consequence of a diabetic intrauterine milieu. More gestational diabetes mellitus is seen in offspring of diabetic mothers than of diabetic fathers (6). Knowler et al. (7) showed that the prevalence of diabetes in children of diabetic mothers is higher when the mother was diabetic during pregnancy than when she developed diabetes after pregnancy.

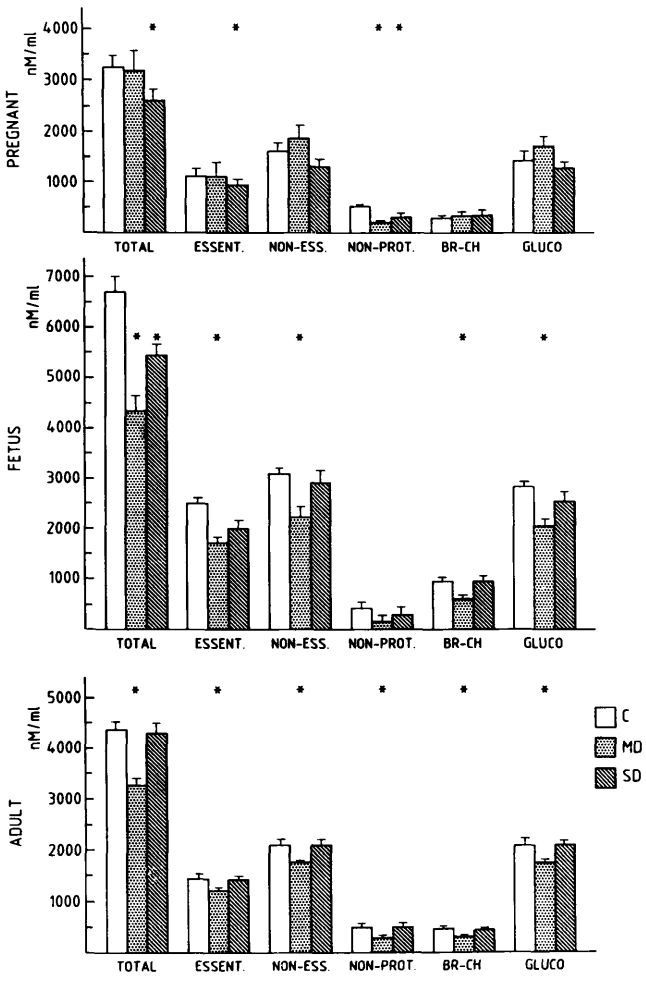

FIG. 2. Amino acid profile in control (C) and mildly diabetic (MD) 1st-generation rats and their fetuses and 2nd-generation adult offspring. For comparison, data of severely diabetic (SD) rats are included. Essent, essential; non-ess, nonessential; non-prot, nonprotein; br-ch, branched chain; gluco, gluconeogenic.

METABOLIC PARAMETERS

To further elucidate the nature of the impaired glucose tolerance in offspring of diabetic rats, we focused on metabolic parameters during experimentally induced diabetes in the first-generation pregnancy and the subsequent generations.

Amino acids are potent stimulators of the fetal endocrine pancreas at the level of development and at the level of β-cell secretion. Changes in the amino acid concentrations in our experimental model might contribute to alterations in the development of the offspring. An amino acid profile was established in nondiabetic and mildly diabetic first-generation pregnant rats and their fetuses and second-generation adult offspring (Fig. 2). There was no difference in the total amino acid level in plasma between nondiabetic rats and mildly diabetic rats at day 20 of gestation. The distribution of the different subgroups of amino acids was similar for all groups. In fetuses of control rats, the total amino acid level in plasma was high. The fetomaternal ratio was 2. On the contrary, in fetuses of mildly diabetic mothers, the plasma amino acid level was lower than in the controls, and the

fetomaternal ratio was 1.3. The low amino acid level in fetuses of mildly diabetic mothers was also present in the subgroups of amino acids. Lower levels of total amino acids and subgroups of amino acids were also present in the adult offspring of mildly diabetic mothers.

From the amino acid data, it is clear that in mild diabetes the level of amino acids in the mother is normal, but the fetal amino acid level is low, so that the fetomaternal ratio is also decreased compared with normal pregnancy. Pregnant rats with mild diabetes offer a normal amount of amino acids to their fetuses. However, the amino acids are utilized by the fetoplacental unit in a different way, suggesting an altered amino acid metabolism. An increased uptake of amino acids by the macrosomic fetuses of mildly diabetic rats was demonstrated by Kim et al. (8). The plasma amino acid level remains lower in the offspring of mildly diabetic rats in the adult state; because the 24-h nitrogen content is increased, it may be proposed that an increased turnover of amino acids is also present in the adult state (9).

GLUCOSE TOLERANCE

In our previous experimental work, we showed that adult offspring of mildly diabetic rats have an impaired glucose tolerance. To further investigate the nature of this intolerance, we submitted adult offspring of control and mildly diabetic mothers to a 3-h continuous glucose infusion. In control rats, the glycemia rose quickly and stayed on a plateau level during the infusion; however, insulin kept rising. In offspring of mildly diabetic mothers, glycemia was higher than normal, whereas insulin levels were lower. The insulin secretory capacity of adult offspring of mildly diabetic rats was reduced. This was confirmed by in vitro studies (10). Basal and stimulated insulin secretion of isolated islets of nondiabetic adults and adult offspring of diabetic rats was measured. After a 90-min incubation of these isolated islets in a high-glucose medium, insulin secretion was markedly decreased in offspring of mildly diabetic mothers.

CONCLUSIONS

These experiments clearly show that even mild diabetes mellitus induces an abnormal intrauterine milieu. The diabetic intrauterine milieu is responsible for the changes in the morphological and functional development of the fetus, with consequences for later life. Mild hyperglycemia of the mother causes fetal hyperglycemia and hyperinsulinemia and an increased utilization of amino acids. In adult life, these animals have a decreased tolerance to a glucose load and keep their activated turnover of amino acids. These changes are induced during intrauterine life and strongly suggest strict metabolic control of the diabetic pregnant woman.

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