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MODULATING EFFECT OF CHOLECALCIFEROL ON VITAMIN D ENDO/PARA/AUTOCRINE SYSTEM IN TYPE 1 DIABETES

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Introduction. Type 1 diabetes (T1D) is a complex autoimmune-endocrine disease that is associated with the development of a number of side effects, one of which is secondary osteoporosis. It is known that the T1D development is accompanied by a significant decrease in the vitamin D bioavailability, which can lead to a disruption of bone remodeling and a decrease in bone mineral density. At present, there is no clear evidence whether disturbances in the insular apparatus and hyperglycemia reduce vitamin D bioavailability, or vitamin D deficiency can indirectly promote the development of T1D. Thus, further investigation is needed to estimate the vitamin D status in relation to the key components of vitamin D endo/para/autocrine system in different tissues of Wistar male rats with experimental T1D and to assess the effects of vitamin D_3 treatment.

Review. The substances of vitamin D group include biologically active sekosteroids vitamin D_3 (cholecalciferol) and D_2 (ergocalciferol). Cholecalciferol hydroxylation in the liver by the enzymes of cytochrome P450 family – CYP27A1 (mitochondrial) and CYP2R1 (microsomal) isoforms of vitamin D 25-hydroxilase, results in the formation of 25-hydroxyvitamin (25OHD), which enters the bloodstream. The next step is the formation of vitamin D-hormone (calcitriol) under the influence of CYP27B1 hydroxylase that occurs either in the kidneys (5%) or in a number of peripheral tissues (95%). Vitamin D catabolism is ensured by CYP24A1, which is responsible for the formation of chemically inert end product – calcitroic acid. Genomic effects of vitamin D require the heterodimmerization of VDR with RXR receptors. VDR-RXR dimmer regulates at transcriptional level the expression of about 500 genes that are responsible for the synthesis of regulatory factors involved in cell proliferation and differentiation, survival, apoptosis and vascular growth.

Vitamin D deficiency and disruptions in the vitamin D endo/para/autocrine system can increase the risk of autoimmune diseases, in particular T1D. Growing evidence suggests that vitamin D plays an important role in preventing autoimmune destruction of pancreatic β-cells and can be directly involved in insulin production, and improvement of the peripheral tissue sensitivity to its action. Different cell types, such as cutaneous and intestinal epithelial cells, macrophages, breast and bone cells, can also express CYP27B1. Additionally, VDR was found in a variety of target cells and its ability to mediate a significant therapeutic potential of vitamin D as a natural VDR ligand in many diseases has been reported.

Type 1 diabetes induces severe impairments of liver and kidneys function that can, at least partially, be ascribed to abnormal hormonal activity of calcitriol. A marked T1D-elicited decline in the circulating 25OHD may result from decreased levels and activity of vitamin D 25- and 25OHD 1α-hydroxylases as well as up-regulation of 24-hydroxylase. Therefore, our aim was to investigate how T1D-associated changes in tissue distribution of CYPs and VDR correlate with vitamin D insufficiency/deficiency and whether vitamin D_3 (cholecalciferol) treatment can affect diabetes-related dysfunction of the vitamin D endo/para/autocrine system.

Results. Type 1 diabetes was induced in male Wistar rats by a single intraperitoneal injection of STZ (55 mg/kg of body weight). It was demonstrated that after 6 weeks of diabetes, the serum level of 25OHD was 50% lower compared with the control. Vitamin D deficiency was accompanied by a significant decrease in the mRNA level of CYP27A1 and CYP2R1 isoforms of 25-hydroxylases in the liver (4.2- and 6.3-fold respectively). The changes in liver vitamin D para/autocrine function were assessed by measuring the expression of mRNA and protein syn-

thesis of CYP27B1 and VDR. We showed a 5.3- and 2.0-fold diabetes-induced decrease in the CYP27B1 mRNA and protein levels, respectively. Diabetic animals also demonstrated a l4.3- and 1.5-fold decrease in the level of VDR, mRNA and protein, vs. control, respectively. In contrast, the level of CYP24A1 mRNA, which facilitates vitamin D catabolism, was 6.4-fold higher in diabetic rats compared with the control.

In the kidneys of diabetic animals, we observed an increase in the levels of both mRNA and protein of CYP27B1 (1.28- and 1.25-fold respectively), and VDR (5.2- and 2.0-fold respectively) in comparison with the control. Moreover, diabetic animals exhibited a 5.0-fold increase in the kidney level of CYP24A1 as compared with the control.

The study of the vitamin D para/autocrine system in bone tissue showed a 13.0-fold increase in the level of CYP27B1 mRNA. In addition, we observed a decrease in both VDR mRNA (3.3-fold) and protein (1.8-fold) in the bone tissue of diabetic animals. Diabetes caused a profound drop in the mRNA level of CYP27B1 (3.8-fold) and VDR (9.0-fold) in the bone marrow as compared with the control. These changes were confirmed by immunofluorescence staining of bone marrow cells followed by confocal microscopy.

Cholecalciferol treatment (600 IU/kg of body weight for 30 days) led to a partial correction of the vitamin D status of diabetic animals that was accompanied by the normalizing effects on the synthesis of the key elements of the vitamin D endo/para/autocrine system in the liver, kidneys, bone tissue and bone marrow.

Discussion. Among various side effects of T1D, impairments in the tissues and organs involved in the metabolism of vitamin D and its hormonal activity through VDR pathway are of particular interest. A decrease in the level of mRNA of 25-hydroxylases in the liver of diabetic animals indicates impairment of the first stage of cholecalciferol activation. Moreover, a decrease in the level of CYP27B1 and VDR indicates the impairments of the para/autocrine function of the liver. This hypothesis is confirmed by an increase in CYP24A1 hydroxylase, which indicates the probable prevalence of vitamin D catabolism over its synthesis in diabetes. An increase in the synthesis of CYP27B1 and VDR in the kidneys of diabetic animals may reflect a compensatory effect of vitamin D endocrine system in response to an overall decrease in blood 25OHD level.

Secondary osteoporosis is recognized as one of the side effects associated with T1D. The elevation of the CYP27B1 mRNA and the down-regulation of the VDR may indicate impairments of osteoblastic/osteoclastic balance with the prevalence of osteoclastogenesis and bone resorption. Vitamin D_3 administered as a potential hydroxylation substrate to form vitamin D-hormone contributes to the complete or partial correction of the most parameters studied.

Conclusions. Diabetes-induced vitamin D deficiency is associated with the abnormalities in renal and extrarenal expression of CYP27B1 and VDR. Vitamin D_3 is effective in amelioration of diabetes-associated impairments of the vitamin D endo/para/autocrine system in different tissues.