

Abstract

Introduction: Diabetes mellitus is a polygene disease and on its manifestation have influence also enviromental factors. We have studied the role of extrapituitary prolactin (PRL) and toll-like receptors (TLR) 2 and 4 in the etiopathogenesis of autoimmune diabetes. PRL is mainly produced by hypophysis, but in small concentrations also in the periphery, where it participates in the immune reactions. Therefore, we investigated the influence of the levels of monocytic PRL mRNA on the development of diabetes, and also the influence of G allele of the -1149 G/T polymorphism in the extrapituitary promotor, which has already been associated with other autoimmune diseases. TLRs are receptors of the immune cells that recognize patogenes entering into the body. They play an important role in the inication of the immune response. We aimed to find out their function in the pathogenesis of the autoimmune diabetes by the detection of their mRNA levels and protein levels expressed on the cell surface of the monocytes.

Material and methods: In this study we included 30 T1D and 21 LADA patients. Three control groups consisted of 23 T2D patients, 23 patients with a nondiabetic disease (neDM) and 60 healthy blood donors (TO). Blood samples have been taken from the individuals. From these blood samples we isolated DNA for genotyping the *PRL* -1149 G/T SNP via RFLP and from monocytes we extracted mRNA for the analysis of the levels of *PRL*, *TLR2* and *TLR4* gene expression via real-time PCR. Further, we separated mononuclear cells that were labelled with antibodies against CD14, TLR2 and TLR4, so that we could detect TLR2 and TLR4 expression on monocytes by using a flow cytometry.

Results: The genotype frequencies of the *PRL* -1149 G/T SNP did not significantly differ between compared groups. Contrary to this, we found statistically significant differences in the levels of PRL and TLR2 mRNA expression between the groups AD and neDM, and in the expression of TLR2 and TLR4 mRNA between AD and TO groups. In the number of monocytes expriming TLRs, we observed higher levels in neDM and TO groups compared to AD. Correlation of the levels of mRNA and proteins in all the individuals demonstrates weak, but statistically significant negative correlation in the monocytes with low expression of CD14 and TLRs.

Conclusion: The *PRL* -1149 G/T SNP does not seem to have any effect on the expresion of the peripheral PRL in monocytes of AD. Patients with autoimmune diabetes have significantly higher levels of TLR2 and TLR4 mRNA compared to healthy controls, but lower numbers of monocytes expriming these receptors on their cell surface. PRL mRNA levels in AD patients were comparable with those in TO group and we conclude that prolactin probably has no influence on the manifestation of autoimmune diabetes.