5. Summary and Conclusion

5.1. Background and Aim

Type 1 diabetes is an autoimmune and chronic inflammatory disease caused by a selective destruction of the insulin producing β-cells in the islets of Langerhans. There are numerous studies showing that proinflammatory cytokines for instance, interleukin IL-1A, IL-B, IFN-γ, and TNF-α are critically involved in the pathogenesis of type 1 diabetes. In this form of diabetes, the rate of β-cell destruction is quite variable, being rapid in some individuals (mainly infants and children) and slow in others (mainly adults) (LADA).

Extracts from the gum resin of Boswellia serrata (BE) and some of its constituents including boswellic acids (BAs) affect the immune system in different ways. Among the various boswellic acids 11-keto-β-boswellic acid (KBA) and acetyl-11-keto-β-boswellic acid (AKBA) have been observed to be active. BAs affect the cellular defence system by interaction with production/release of cytokines. Thus, BAs inhibit activation of NFκB which is a product of neutrophile granulocytes. Consequently downregulation of TNF-α and decrease of IL-1, IL-2, IL-4, IL-6 and IFN-γ, which are proinflammatory cytokines by BEs and BAs has been reported. In the same context the inhibitory action of BAs on 5-LO leading to a decreased production of leukotrienes play an important role in treatment of variety of chronic inflammatory diseases which associated with increased leukotriene activity.

This study was conducted in order to examine whether or not Boswellia serrata extract, KBA or AKBA could prevent or at least reduce pancreatic insulitis, periinsular apoptosis cytokines as well as hyperglycemia induced by MLD-STZ which is an acknowledged model for human type 1 diabetes.

5.2. Methods

5.2.1. Standardization of Boswellia serrata extract

Boswellia serrata extract was standardized by HPLC method to determine the exact amount of KBA and AKBA in our extract. Our extract was containing KBA (5.48%) and AKBA (4.66%).

5.2.2. Induction of Type 1 Diabetes

Mice were injected with STZ 40mg/kg for 5 consecutive days according to a protocol of MLD-STZ which induce hyperglycemia through an autoimmune process.

5.2.3. Blood glucose measurement

Blood glucose was measured at days 0, 5, 10, 14, 21, 28 and 35. Blood samples were taken from mouse tail and measured by mean of Accu-Chek aviva glucometer.

5.2.4. Histology and Immunohistochemistry

Pancreases were isolated at day 35 for H&E staining and at day 10 for immunohistochemistry staining (CD3-staining and caspasse-3 staining). Therefore, all pancreases were carefully isolated and immediately fixed and kept in buffered 4% formalin.

5.2.5. Determination of Serum Cytokines

Different types of cytokines (G-CSF, GM-CSF), proinflammatory cytokines (IL-1A, IL-1B, IL-2, IL-6, IFN-γ and TNF-α) and anti-inflammatory cytokines (IL-4 and IL-10) were determined in the serum of mice by ELISA method.

5.3. Results

- Administration of STZ for 5 successive days increased blood glucose levels, caused infiltration of lymphocytes into pancreatic islets and produced apoptosis of periinsular tissues. These effects were associated with increase of colony stimulating factors (G-CSF, GM-CSF), proinflammatory cytokines (IL-1A, IL-1B, IL-2, IL-6, IFN-γ and TNF-α) and anti-inflammatory cytokines (IL-4 and IL-10).
- Simultaneous administration of BE, KBA or AKBA prevented the increase in blood glucose levels in STZ-treated mice
- BE as such did not change blood glucose levels in control mice.

- BE, KBA and AKBA prevented the infiltration of lymphocytes into pancreatic islets and apoptosis of periinsular tissues.
- BE, KBA and AKBA also prevented increases of colony stimulating factors (G-CSF, GM-CSF), proinflammatory cytokines (IL-1A, IL-1B, IL-2, IL-6, IFN-γ and TNF-α) and anti-inflammatory cytokines (IL-4 and IL-10).

5.4. Conclusions

From our results we conclude that:

- BE as well as its main active constituents KBA and AKBA could prevent or at least reduce insulitis and apoptosis of pancreatic islets of MLD-STZ diabetic mice.
- Consequently they prevented the elevation of blood glucose level.
- These effects may be due to their inhibitory action on the release of proinflammatory cytokines.
- Since BE, KBA and AKBA prevent insulitis caused by proinflammatory cytokines in the model of MLD-STZ diabetes, it is conceivable that BE and BA may also prevent or treat insulitis in type 1 diabetes and LADA.
- However, clinical studies are necessary to prove this hypothesis in Humans.