Type 1 diabetes is an autoimmune disease associated with pancreatic Langerhans beta-cells destruction, resulting in a lack of endogenous insulin, impaired glucose homeostasis, and a decrease in the quality of patient’s life. The etiology of the disease is still not well established due to the inaccessibility of the affected endocrine pancreatic beta-cells. The insight into the physiological processes in the affected beta-cells can be achieved with the study of extracellular vesicles, which can be biomarkers and mediators of the disease development. In our study, a fraction of blood plasma extracellular vesicles positive for membrane proteins potentially associated with insulin-producing beta-cells was observed, which indicates beta-cell communication with other tissues and the immune system. The next-generation sequencing was used to analyze blood plasma extracellular vesicle RNA-fractions of type 1 diabetes individuals and intensive beta-cell destruction in Langerhans islets transplantation. The immunomodulatory role of the selected differentially expressed extracellular vesicle miRNAs was evaluated with the whole human blood samples in vitro stimulation, which resulted in vesicle-miRNAs accumulation in phagocytes endolysosomal pathway and the activation of the immune system. The immune system activation presented with the increased degranulation and proliferation of NK and T-cells, as well as increased cytokine/chemokine release, while the miRNA transfection together with the chloroquine inhibitor resulted in the decreased inhibition of the immune system response. The chloroquine inhibition indicated TLR7/8 responsible for vesicle delivered miRNA recognition. Vesicle delivered miRNAs in type 1 diabetes show the complexity of extracellular vesicles RNA in the regulation of the immune system and a potential involvement in the development of autoimmunity. The TLR7/8 signaling also emphasizes the implications for developing strategies for disease prevention.