

in Prague, 3.9. 2024

Supervisor's report on diploma thesis by <u>Bc. Anna Zelenska</u> "Effect of low-carbohydrate diet on development of type 1 diabetes and immune parameters in NOD mice"

This diploma thesis has been worked out by Bc. Anna Zelenska in the Laboratory of Cellular and Molecular Immunology, IMIC, CAS, Prague, as part of a project investigating role of Low-carbohydrate diet (LCD) on glycemic control and islet autoimmunity in type 1 diabetes (T1D).

Bc. Anna Zelenska carried out experiments focused on effects of a low-carbohydrate diet on the development of T1D and immune parameters in the NOD mouse model if applied at different age. This diploma thesis brings some preliminary data on the effects of the LCD if applied at adult age of 4 weeks and also unexpected data on the effect of LC diet if introduced early in life, already to the pregnant females. The thesis is well written, aims are concise, and a great deal of flow cytometry data, although preliminary, is included in the results section. Discussion is pointing to possible mechanisms of the unexpected acceleration of diabetes and immune changes in already high-diabetes incidence colony of NOD mice.

Bc. Anna Zelenska has learned several new methods in the lab that are presented in her thesis. Her approach to experimental work is very careful and precise. She has also easily and critically worked with the relevant literature.

Her project was negatively influenced by the closure of the animal facility at the IMIC (during 2023) and a later start of our new animal facility, that has become fully operational from since the beginning of 2024. Despite this, she managed to test the diet at two time-points and repeated the unexpected results several times, as well as participated and carried out and several flow cytometry experiments. The unexpected results presented in her diploma thesis will occupy our lab for much longer time.

In conclusion, Bc. Anna Zelenska has learned several new methods that she presents in her diploma thesis. She has carried out the animal experiments with great care, gained new data, contributed to the experimental design, and easily and critically worked with the relevant literature. That is why, I can definitely recommend her thesis for the opposition proceedings and for the awarding the Master's degree title.

David Funda, M.D., Ph.D.

Ind Fent

Laboratory of Cellular and Molecular Immunology,

IMIC AS CR, v.v.i.