

Preventing injuries during the training is critical to optimize performance, and physical exercise may be associated with the induction of both acute and chronic inflammatory changes associated with excessive reactivity of the immune system and repeated tissue damage. However most biomarkers are insufficient and show only modest association with susceptibility to tissue injury. Latest advances in molecular biology, immunohistochemistry, bioinformatics and computer engineering allowed for a highly comprehensive insight into dynamic bacterial consortia and its' reflection in the immune system. Given the gut microbiota's (so the microbe population living in our intestine) fundamental role in the regulation of energy metabolism, inflammatory response, digestion, nutrition, hydration and oxidative stress, we are convinced about the great role of understanding how the gut microbiome may exert beneficial effects on the training people and whether changes in its composition are a limiting or supportive training factor.

Although, there are scientific reports confirming substantial role of gut microbiota in health maintaining and better results in the field of sport, as well as highlighting the greater diversity of microbiome after severe injury, majority of them are based on an animal model and the results are inconsistent. For example, study published in 2013 on 2 mice groups showed that the exercise increased phylum *Firmicutes*, class *Bacilli* and most of these were in the order *Lactobacillales*. The other work conducted in 2014 on mice, revealed at the phyla level, that the exercise reduced *Bacteroidetes*, while increased *Firmicutes*, *Proteobacteria* and *Actinobacteria*. Although, the results of GM analysis performed on different models are inconsistent, majority of studies revealed that after a few weeks of endurance exercise, the level of *Firmicutes* increased, while the level of *Bacteroidetes* decreased, what was also related to endurance athletes.

Additionally it has been already observed that under muscular stress, changes in iron metabolism occur. Iron is one of the most important factors necessary for bacterial growth, therefore changes in host iron homeostasis may affect iron content in the intestine, thereby composition of gut microbiome. That was confirmed by research on the mouse model, where *IRP2* gene, responsible for iron metabolism was modified, which contributed to a significant change in the intestinal microbiome. However the effect of exercise on the human gut microbiota has not been investigated in the context of muscle performance and iron metabolism. Alike an association between body fat and muscle concentration and gut microbiota in context of inflammatory response to physical activity (in which the change in free iron concentration is significant), has also not been explored. However, in some studies it has been shown that an obesity-associate GM can significantly contribute to host weight gain through enhanced energy harvest and drive inflammation through reduced gut barrier integrity. This relates to changes in hepcidin, ferritin, transferrin and free iron concentration. It can affect diverse body's response to physical exercise, as well as the ability to achieve good results in sport.

Because there is still lack of knowledge regarding to the correlation of human gut microbiota composition with clinical features related to the soft tissue injuries occurring after training depending on its' intensity, we would like to focus mainly on the issue of intestinal flora variability in the context of many years of training adaptations in sports. The main aim of proposed project is detailed and combined characteristics of bacteria affecting those features by using novel next generation sequencing technique (NGS). A substantial part of our project is dedicated to define a so-called „window of opportunity” that in this context means the time when the crosstalk between bacteria and answer of the immune system to the specific type of injury is the most crucial and can be traced. Because there are some indications that GM may be modified by cfDNA, which substantially increases during i.e. tissue damage, we would like to explore those correlations more deeply in terms of the type of physical activity and tissue injury. This can contribute to finding new, useful tools in predicting the occurrence of sports injuries. Furthermore, the knowledge about changes in iron metabolism can give us an answer on the role of GM in sport and physical activity in the context of iron homeostasis in the body. Moreover, it may lead to an answer how iron changes can influence human microbiome and whether there is a direct relationship between the iron state and the human microbiome in both - the active and non-active population. This may serve also to identify species helpful for athletic achievement in context of iron status and microbiome state. In addition to giving us a better understanding of how a healthy lifestyle shapes the gut microbiota, these studies could lead to potential treatments or probiotics for ailments or potentially improving athletic performance and reduce the risk of soft tissue injuries.