

Betaine is a chemical compound which can be found naturally in food, including wheat bread, spinach, and beets. Betaine can act as an organic osmolyte to protect cells under stress or it can be a source of methyl groups in many biochemical pathways. Betaine intake can stimulate muscle growth and decrease in body fatness, but it can also lead to changes in lipid metabolism. For this reason betaine is used in animal feed and supplements for athletes. Molecular mechanisms of betaine action are not fully understood.

The main aim of the project is thus to analyze several potential mechanisms through which betaine can affect metabolism. We suppose that betaine can regulate lipogenesis, gluconeogenesis,  $\beta$ -oxidation of fatty acids, fatty acid transport, functioning of white adipose tissue (WAT) or biogenesis of mitochondria. We also hypothesize that betaine can regulate insulin-like growth factor 1 (IGF-1) or AMP-activated protein kinase (AMPK) signaling and thereby muscle growth.

Our study includes an in vitro and experiments on laboratory rodents. The purpose of the cell culture experiment is to analyze AMPK and IGF-1 signaling in liver and muscle cells after betaine supplementation. The experiments on animals were designed to compare the effects of betaine supplementation in sedentary and exercising rats. Two situations will be considered: effects of betaine in normal-weight rats and in rats with a high-fat diet induced obesity. At the beginning and at the end of the experiments body composition and gas calorimetry measurements (e.g. oxygen consumption and energy expenditure) will be performed. To analyze metabolic effects of betaine supplementation we will be measuring homocysteine, fasting glucose, total cholesterol, HDL and LDL cholesterol, and triacylglycerol. Betaine, carnitine, and methionine concentrations will be measured in blood, liver, and muscle. Moreover, activity of genes involved in regulation of metabolism will be measured in the liver, muscle, and WAT. Additionally, phosphorylation (activity) of proteins involved in AMPK and IGF1 signaling will also be analyzed. Finally, histological analyzes of tissues will be performed.

Our project can significantly broaden basic knowledge on regulation of metabolism. Additionally, as betaine supplementation has gained much attention in athletes and there are still some doubts on its safety, our in depth study can also have a practical meaning.