Abstract

Obesity, one of the most serious health problems of the 21st century, often occurs as a result of an imbalance between energy intake and energy expenditure. Dietary lipids play an important role in the development of obesity, partly because they represent the richest source of energy amongst all macronutrients. It is, however, not only the amount of consumed lipids, but also the composition of fatty acids, which strongly influences health effects of a particular diet. Saturated fatty acids (SFA) are generally considered as unhealthy due to their pro-inflammatory and lipotoxic properties, while monounsaturated fatty acids (MUFA) and polyunsaturated fatty acids (PUFA) represent a healthier alternative, as they are more readily oxidized and do not disrupt biochemical properties of cellular membranes. Amongst PUFA, PUFA of *n-3* series (Omega-3) represent an utterly unique class of lipids that have been documented to protect against cardiovascular disease and dyslipidemia in men and improve insulin sensitivity and glucose tolerance primarily in animal models of obesity.

Some molecular mechanisms of Omega-3 action have been already uncovered, such as the modification of biological membranes composition, activation of various transcription factors and membrane receptors, and their role as precursors for the synthesis of bioactive lipid mediators with anti-inflammatory and pro-resolving properties. In this thesis, we explored novel mechanisms and systems, which may be influenced by long-term dietary supplementation of Omega-3 to male C57BL/6 mice with obesity induced by a high-fat diet rich in PUFA of *n-6* series (Omega-6). Furthermore, we compared the effects of Omega-3 administered either in the form of triacylglycerols (Omega-3 TAG) or marine Omega-3 phospholipids (Omega-3 PL); the latter form of Omega-3 has been recently shown to induce more potent and consistent metabolic effects, especially on glucose metabolism and liver fat accumulation (i.e. hepatic steatosis), and, therefore, could represent a better alternative regarding the prevention and potentially also the treatment of obesity-linked metabolic diseases.

In the first project (Publication A), we analyzed the effects of Omega-3 TAG depending on the type of other lipids in the diet, i.e. the diets with the prevalence of either Omega-6 (cHF diet) or SFA and MUFA (LHF diet). Without Omega-3 supplementation, the two diets *per se* did not differ in their impact on body weight, the amount of body fat (adiposity) or insulin sensitivity examined by the hyperinsulinemic-euglycemic clamp technique. It could be due to the protective up-regulation of the enzyme stearoyl-CoA desaturase 1 (SCD1) that converts potentially harmful SFA contained in the LHF diet into less toxic MUFA. The accumulation of MUFA, however, might be connected to more pronounced hepatic steatosis, which was typical for LHF-fed animals. Omega-3 supplementation ameliorated hepatic steatosis by repressing lipogenesis and promoting fat oxidation, and these effects were independent of other lipids in the diet. On the contrary, white adipose tissue (WAT) inflammation and glucose tolerance were beneficially affected by Omega-3 only when supplemented within the cHF background, while these parameters tended to deteriorate further when Omega-3 were present in the LHF diet. The results of this project suggested

that the supplementation of Omega-3 on SFA-rich dietary background could be problematic, since under these conditions Omega-3 supplementation interferes with the protective mechanism based on the increased activity of SCD-1 in response to LHF feeding.

The second project (**Publication B**) was based on the observation that the long-term supplementation of Omega-3 TAG in dietary obese mice leads to increased plasma insulin levels following glucose administration, but only when glucose is applied orally; it was predicted that increased activity of the incretin system could be involved. We were unable to prove that Omega-3 supplementation increased the activity of the incretin system, as neither an increased secretion of glucagon-like peptide-1 (**GLP-1**), decreased activity of the degrading enzyme dipeptidyl peptidase 4 (**DPP-4**), nor changes in sensitivity towards glucose-dependent insulinotropic polypeptide (**GIP**) were observed following Omega-3 intake. However, Omega-3 supplementation normalized hypersecretion of GIP and increased concentrations of GIP receptors in WAT, which was otherwise observed in obese mice. Thus, the above effects of Omega-3 on GIP may represent a novel pathway by which these fatty acids could affect adiposity.

Finally, in **Publication C** and **Publication D**, we focused on the long-term dietary supplementation with Omega-3 PL derived from either herring meal or Krill oil. We showed that Omega-3 PL were able to efficiently reduce hepatic steatosis and normalize dyslipidemia by a complex and integrated inhibition of *de novo* lipogenesis and cholesterol biosynthesis together with stimulation of the oxidation of fatty acids, observed at the level of hepatic gene expression, while the presence of both Omega-3 and their lipid carrier, i.e. phospholipids, namely phosphatidylcholine (**PC**), are necessary to achieve the maximum effect. Furthermore, by using the hyperinsulinemic-euglycemic clamps in obese mice, we showed that Omega-3 PL ameliorated obesity-linked glucose intolerance and whole-body insulin resistance, mainly due to the beneficial effects on hepatic and muscle insulin sensitivity, while this effect was superior to Omega-3 TAG administered at a comparable dose.