Abstract

Obesity is a serious problem in society today [1,2]. It might seems to have been caused simply by excess consumption of food compared to energy expenditure but obesity is actually a complex metabolic disorder centred on adipose lipid metabolism and cellular signalling systems linked to it [3]. Understanding the biology of adipose tissue (AT) is very important for the identification of novel and potential therapeutic targets in order to prevent and treat obesity-related disorders [4]. We utilized analytical approach liquid chromatography coupled to mass spectrometry (LC-MS) to study adipose tissue metabolism. Also, we were especially interested in the effect of omega-3 polyunsaturated fatty acids (PUFA) on that metabolism. Rodent and cell line experiments were performed and analyses were done of white adipose tissue (WAT), serum/plasma samples or cells as well as milk samples from mothers.

At first, we established several ultra performance liquid chromatography-tandem mass spectrometry (UPLC-MS/MS) methods for analysis of acylcarnitines (AC), amino acids (AmA) and other metabolites. Importantly, these methods were able to distinguish isobaric species of AC which is not usually possible. Using these approaches we uncovered several acylcarnitines, i.e. long chain AC, carnitine, acylcarnitine C4 and C6, C4DC, as well as amino acids tyrosine, alanine, ornithine and threonine that could serve as complex, gender-specific biomarkers of propensity to obesity and partially as a biomarker of obesity-associated insulin resistance. It was discovered that maternal intake of a cafeteria diet during lactation in rats alters in the plasma profile of AC and AmA. Specifically, it was noticeable in acetylcarnitine and medium- and long-chain AC, as well as glycine, alanine, isoleucine, serine and proline, in the offspring.

Second, we focused on mutual interaction between two main cell types of adipose tissue, adipocytes and adipose tissue macrophages. Our results showed that macrophages modulate lipolysis and fatty acid re-esterification in adipocytes while their modulation depends on polarization states of macrophages and responses to dietary omega-3 PUFA supplementation. Then UPLC-MS/MS metabolomic method revealed that inflammatory M1 and anti-inflammatory M2 macrophages act differently. This thesis also explores lipid mediators synthesized in adipose tissue. As a result, the origin of lipid mediator protectin D1 was discovered. Also, levels of several lipid mediators in different cell types of WAT were determined and showed interplay between them. Moreover, we documented the omega-3 PUFA´s ability to lower the inflammation in WAT.

Later on, we concentrated on branched fatty acid hydroxy fatty acids (FAHFA) lipid molecules that influence WAT metabolism as well. We identified a novel compound of FAHFA lipid class, docosahexaenoic acid hydroxylinoleic acid (DHAHLA), by its fragmentation scheme and detected several DHAHLA´s isomers in murine serum. We then demonstrated that adipocytes and macrophages are able to synthetize 13-DHAHLA and we proved its anti-inflammatory and pro-resolving properties. Subsequently, we found 13-DHAHLA in human breast milk for the first time but only in samples from mothers who were supplemented with omega-3 PUFA during pregnancy. It is also important to indicate that FAHFA from breast milk reach a newborn's circulation and thus potentially positively influence its metabolism.

In summary, we used very sensitive UPLC-MS/MS methodology to broaden our knowledge on adipose tissue metabolism. Specifically, we focused on mutual interaction between main components of adipose tissue and we analysed different metabolites (e.g. acylcarnitines, lipid mediators, FAHFA compounds) that influence adipose tissue metabolism in various samples.