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Studies on LXR and CIDEA function in human adipocytes

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ABSTRACT

Obesity, defined as a body mass index of 30 or above, is the result of an imbalance of energy intake and energy expenditure. In the last decade, the obesity prevalence has truly reached epidemic proportions with major effects on public health. Obesity is closely associated with insulin resistance, type 2 diabetes, hyperlipidemia and atherosclerosis. On the other end of the spectra, cancer cachexia is a poor diagnostic factor in cancer patients. It is characterized by a state of unintentional weight loss, primarily of body fat but also of lean body mass. Although the mechanisms behind the loss of adipose tissue are not completely understood, lipolysis seems to be a major factor.

Adipose tissue is an important metabolic and endocrine organ. One of the most important functions of the adipocyte is lipolysis, the hydrolysis of triglycerides to free fatty acids (FFA) and glycerol. This is a tightly regulated process of great importance to whole-body metabolism. The FFAs can either be released into the circulation, to be used as energy substrate by other organs and tissues, or utilized within the adipocyte for re-esterification or lipid oxidation. The process of lipid oxidation in adipocytes is controlled in part by the pyruvate dehydrogenase complex (PDC), an important regulator of substrate oxidation in adipocytes. This complex is inactivated when phosphorylated by pyruvate dehydrogenase kinases (PDKs), which promotes lipid oxidation rather than glucose oxidation.

The aim of this thesis was to investigate how two factors, the liver x receptor (LXR) and cell death-inducing DNA fragmentation factor-α-like effector A (CIDEA) affect adipose tissue metabolism. LXR is a nuclear receptor and a known regulator of cholesterol, lipid and carbohydrate metabolism. CIDEA is almost exclusively expressed in white adipocytes in humans and can affect critical metabolic functions such as lipolysis.

In **paper I**, we investigated the role of CIDEA in cancer cachexia. We measured levels of CIDEA in subcutaneous adipose tissue from subjects suffering from cancer cachexia and compared these to weight-stable cancer patients and noncancer patients. Levels of CIDEA mRNA were increased in cancer cachexia and correlated with elevated levels of FFAs and weight loss. Over-expression of CIDEA increased fatty acid oxidation in human adipocytes in culture and decreased glucose oxidation. Furthermore, augmented levels of CIDEA enhanced the expression of PDK1 and PDK4, and the phosphorylation of PDC. In accordance with this, mRNA levels of PDK1 and PDK4 in the clinical material correlated with CIDEA expression. In conclusion, CIDEA is involved in loss of adipose tissue in cancer cachexia at least in part due to its ability to inactivate PDC and thereby switch substrate oxidation in human adipocytes from glucose to lipids.
In **papers II and III**, the role of LXR in human adipocytes was studied, with focus on substrate oxidation (paper II) and lipolysis (paper III). In paper II, we treated human adipocytes with the LXR agonist GW3965 and observed an increased fatty acid and decreased glucose oxidation. We showed that LXR activation can increase the mRNA level of PDK4 and thereby the phosphorylation of PDC. We also showed a decreased activity of PDC, which was found to be dependent on PDK4. Furthermore, we could establish that the effect of GW3965 on lipid oxidation was specific for LXR, since it was abolished upon knockdown of LXR. In conclusion, we suggest that LXR has an important role in the regulation of substrate oxidation in human adipocytes, at least in part by influencing the phosphorylation status of PDC.
In paper III, LXR activation was shown to up-regulate glycerol release from human adipocytes. Based on microarray analysis we found a strong impact of LXR activation on known lipolysis-regulating genes. We showed differences in expression and localization of perilipin 1 (PLIN1) and hormone sensitive lipase (HSL). When PLIN1 is depleted, the effect of LXR is abolished. Furthermore, we showed binding of LXR and its heterodimerizing partner Retinoid X Receptor to the promoters of HSL and PLIN1 upon LXR activation. We also demonstrated that LXRα is the predominant isoform involved in regulation of adipocyte lipolysis within this context. In conclusion, we proposed that LXR activation up-regulates adipocyte lipolysis, at least in part through LXR binding to the promoter of PLIN1 and down-regulation of PLIN1 expression.

In conclusion, we suggest that CIDEA and LXR can affect central functions of adipocyte metabolism, namely lipolysis and substrate oxidation. We show that CIDEA is involved in the loss of adipose tissue in cancer cachexia and that this is at least in part due to a shift in substrate oxidation. Activation of LXR in human adipocytes increases fatty acid oxidation and lipolysis, through effects on PDC and PLIN1. The findings in this thesis are of importance for conditions of dysregulated adipose tissue metabolism, such as obesity and cachexia.