Neuregulin 4: A "Hotline" Between Brown Fat and Liver

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The discovery that functional brown adipose tissue (BAT) in adult humans is inversely related to body fat mass and may reflect metabolic health has stimulated adipose tissue research to explore activation of BAT as a potential target for antiobesity treatments. In addition to the capacity of BAT to increase energy expenditure and glucose and lipid uptake, BAT secretes factors that may contribute to the regulation of whole-body metabolism. Among signals released from BAT, neuregulin 4 (NRG4) has been recently identified as an endocrine factor that may link the activation of BAT to protection against diet-induced obesity, insulin resistance, and hepatic steatosis. NRG4 was shown to directly reduce lipogenesis in hepatocytes, and it could indirectly activate BAT via sympathetic neurons or via inducing brown adipocyte–like signatures in white adipocytes in a paracrine manner. However, the potential relevance of NRG4 as a diagnostic tool or target for the treatment of obesity-related diseases remains to be explored.

Brown Adipose Tissue: An Emerging Target to Treat Obesity and Metabolic Diseases?

In the past decade, brown adipose tissue (BAT) thermogenesis has emerged as a potential treatment target for antiobesity strategies. Although increasing energy expenditure through cold-induced or pharmacological activation of BAT could theoretically contribute to weight loss, so far there is no evidence that BAT stimulation or recruitment can be used for therapeutic weight loss in humans (1). In addition, the extent of BAT activation as a clinically relevant target for body weight loss is limited by the potential cardiovascular risk, including increased heart rate and blood pressure (1). On the other hand, improvements in whole-body insulin sensitivity and glucose and lipid metabolism have been reported upon cold-induced BAT activation in humans (2). These beneficial metabolic effects have been attributed to BAT-dependent thermogenesis, which is mediated through the uncoupling of mitochondrial fuel oxidation from ATP production by the expression of uncoupling protein 1.

Signals from BAT

More recently, it has been recognized that molecules secreted or released by BAT ("BATokines") may contribute to the systemic effects of BAT activation (3). In an autocrine and paracrine manner, BAT-secreted factors mediate the adaptation of BAT to stimuli, such as cold, by promoting hypertrophy, hyperplasia, vascularization, innervation, and blood flow of BAT (3). In addition, molecules released from BAT can act as endocrine factors on target organs (including the liver, pancreas, and heart), bone, and the nervous system. The secretory proteome of brown adipocytes detected in the cell culture medium could be distinguished into proteins that are either upregulated (e.g., adiponectin, fatty acid–binding protein 4, retinol-binding protein 4, adipsin, chemerin) or downregulated (e.g., clusterin, apolipoprotein E, macrophage migration inhibitory factor) in response to cyclic AMP–mediated thermogenic activation (4).

Signals released from BAT further include adenosine, nitric oxide, triiodothyronine, fibroblast growth factor 21, interleukin-6, and neuregulin 4 (NRG4) (3,5). These factors may represent the mechanistic link underlying the association between BAT activation, protection against obesity, and associated metabolic alterations.

Role of NRG4 in Obesity-Related Diseases

NRG4 has been identified as a member of the epidermal growth factor family, which is most highly expressed in BAT of mice (6,7). White adipose tissue in both mice and humans expresses NRG4, whereas NRG4 expression is very low in skeletal muscle, heart, liver, and brain tissue (7). Neuregulins represent a distinct subgroup of growth factors that contain an epidermal growth factor–like domain and primarily signal through the receptor tyrosine-protein kinases/human epidermal growth factor receptors (ErB/HER) ErbB3 and ErbB4 to regulate diverse biological processes (6). NRG4 is synthesized as a transmembrane precursor and undergoes proteolytic cleavage (7). The resulting extracellular fragments are released and are biologically active on ErbB4-expressing target cells (7).

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Recently, NRG4 has gained attention as a potential novel target for the treatment of obesity-associated disorders, including type 2 diabetes and nonalcoholic fatty liver disease (7). For example, low serum NRG4 concentrations are associated with obesity, fatty liver disease, and insulin resistance in children with obesity (8). Wang et al. (7) showed that NRG4 expression was highly enriched in BAT, that NRG4 expression is increased during brown adipocyte differentiation, and that NRG4 expression is lower in adipose tissue of rodent models and humans with obesity (7). Moreover, higher NRG4 expression was found in murine white adipocytes acquiring a brown phenotype (beige or brite adipocytes) in response to cold exposure (9).

Supporting a mechanistic role of NRG4 in the development of obesity, Nrg4-deficient mice (Nrg4$^{-/-}$) gained significantly more body weight and developed more pronounced hepatic steatosis and insulin resistance, compared with controls, upon high-fat feeding (7). In contrast to the loss-of-function studies in Nrg4$^{-/-}$ mice, overexpression of NRG4 in adipose tissue leads to less fat accumulation (including less liver fat), improved glucose tolerance, and insulin sensitivity (7).

In a cohort of 642 individuals with a wide range of body fat mass and metabolic parameters, we found significant inverse correlations between subcutaneous adipose tissue *NRG4* mRNA and whole-body fat as well as liver fat (7). Moreover, lower adipose tissue NRG4 expression was associated with impaired glucose tolerance or type 2 diabetes (7).

NRG4 Links BAT to Liver

Specific binding assays on sections from different tissues identified the liver as the main NRG4 target tissue (7). NRG4 effects on the liver include both metabolic regulation and protection of hepatocytes from dietary stress–induced injury (10) (Figure 1). Reduced NRG4 function was associated with a significantly higher expression of genes involved in de novo lipogenesis in the liver, whereas NRG4 overexpression caused reduced hepatic expression of lipogenic genes (7). In addition, NRG4 signaling in the liver may play a role in the progression from liver steatosis to nonalcoholic steatohepatitis (NASH) (10). In a mouse model for diet-induced NASH, NRG4 deficiency was associated with accelerated liver injury, fibrosis, and inflammation, whereas NRG4 overexpression in adipose tissue reduced diet-induced NASH (10). Guo et al. (10) showed that hepatic NRG4 signaling protected against diet-induced NASH by activating a cellular survival pathway.

Role of NRG4 in Neurite Outgrowth

In addition to effects on the liver, NRG4 might also target other tissues and cells (5). In this context, it has been suggested that adipocyte-derived NRG4 induces neurite outgrowth and could thereby regulate activation of BAT by sympathetic neurons (5,9) (Figure 1). Bone morphogenetic protein 8b may play an important role as an activator of NRG4 secretion and as an interconnected regulator (11). In addition, effects of NRG4 on the liver, but also on BAT and white adipose tissue,
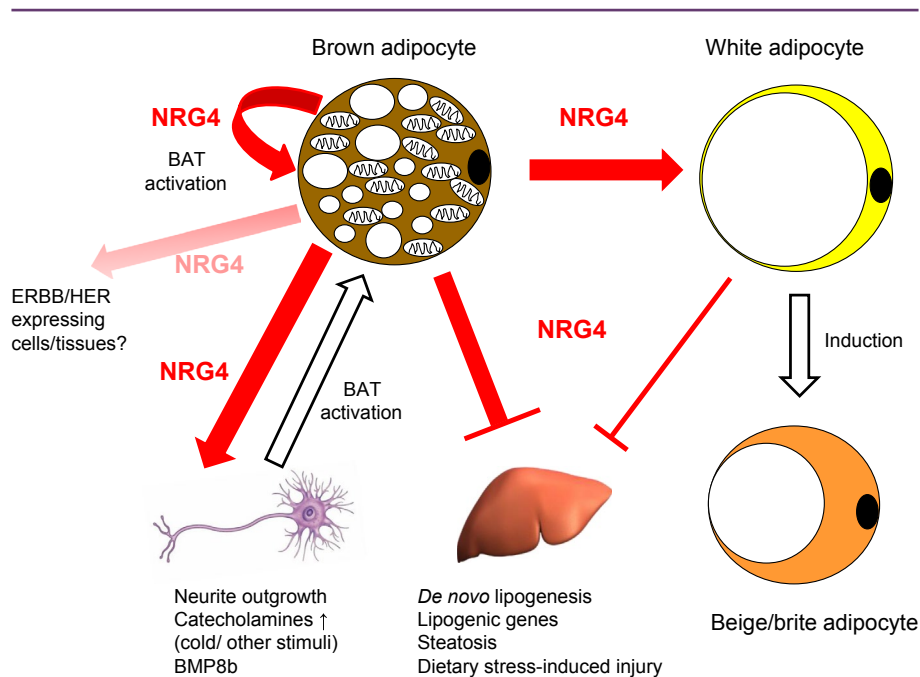

Figure 1 Effects of neuregulin 4 (NRG4) secretion from brown and white adipocytes on target tissues. NRG4 has higher expression in brown compared with white adipocytes. NRG4 could, in an autocrine/paracrine manner, regulate adipose tissue function. NRG4 secretion from adipocytes may stimulate outgrowth of neurites from sympathetic neurons, which in turn may contribute to activation of brown adipose tissue (BAT) through cold-induced (or by other stimuli) catecholamine release. This process is facilitated by bone morphogenetic protein 8b (BMP8b). NRG4 secretion inhibits hepatic de novo lipogenesis and expression of genes involved in lipogenesis in the liver. NRG4 could facilitate the induction of beige/brite from white adipocytes. Most likely, NRG4 exerts its effects on other target cells and tissues expressing ErbB/HER receptors. The figure was modified from Pfeifer (5) and is based on data from Wang et al. (7), Rosell et al. (9), Guo et al. (10), and Pellegrinelli et al. (11).

may be related to increased sympathetic innervation, which drives neurovascular remodeling in these tissues (11). Whether increased innervation mediated by NRG4 also contributes to the observed metabolic effects and may lead to increased lipolysis has not been directly investigated.

The Potential of NRG4 as a Drug Target

Collectively, human data and the phenotype of mice with either a gain or loss of NRG4 function suggest that reduced NRG4 may be causally linked to obesity-related fatty liver disease and impaired glucose metabolism. The exact mechanisms of how NRG4 exerts these beneficial effects are not entirely clear but may involve direct effects of NRG4 (as an endocrine signal) on hepatocytes to reduce lipogenesis and diet-induced liver injury, on indirect activation of BAT via sympathetic neurons, or on inducing brown adipocyte–like signatures in white adipocytes in an autocrine/paracrine manner (Figure 1).

The potential of NRG4 to become a target for the pharmacotherapy of obesity and related diseases, including type 2 diabetes and nonalcoholic fatty liver disease, will also depend on possible "off-target" effects. ErB/HER receptors regulate key processes, including cell proliferation, survival, and differentiation, and play an important role in cancer cell growth and survival (6). It has been shown that recombinant NRG4 induces tyrosine phosphorylation of ErbB4/HER4 and activates the proliferation of lymphoma cell lines (12). However, there is no direct evidence that activation of NRG4/ErbB4 signaling promotes cancer development. On the contrary, Liu et al. (13) demonstrated that loss of ErbB4 contributed to the development of hepatocellular carcinoma and suggested *ErbB4* as a tumor suppressor gene.

In summary, NRG4 underlines the importance of BAT secretory function in the context of obesity and metabolic disorders. Whether NRG4 may become clinically relevant as a marker of metabolic health or as a target for the treatment of metabolic diseases needs to be further explored.**O**

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