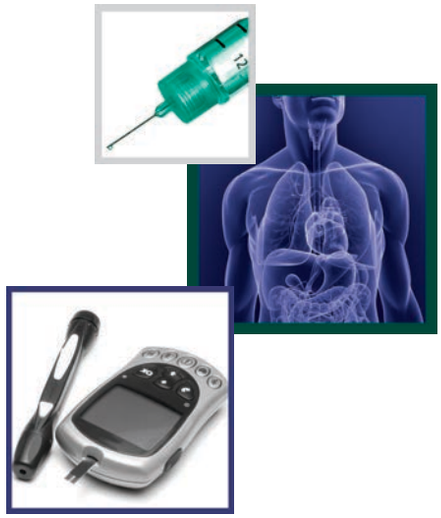
REVIEW



Importance of adipokines in glucose homeostasis

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Practice Points

- Adipose tissue is an active endocrine organ secreting hundreds of molecules; so called adipokines.
- Adipose tissue dysfunction may represent an early defect in obesity, which is characterized by the secretion of a diabetogenic, proinflammatory and atherogenic adipokine pattern.
- Circulating adipokines may cause and reflect alterations in glucose metabolism.
- Clinical relevance of adipokines includes their use both as biomarkers for adipose tissue function and distribution, and as potential treatment of obesity-related diseases.
- Adiponectin may improve insulin sensitivity.
- Leptin treatment improves insulin sensitivity and lowers serum concentrations of triglycerides in patients with lipodystrophy.
- Leptin can be used as a replacement treatment for patients with inherited leptin deficiency.
- Chemerin and RBP4 are biomarkers for insulin resistance and visceral fat distribution.
- High expression and activity of DPP-4 is associated with abdominal obesity.
- Administration of apelin and vaspin improves glucose metabolism in experimental animals.

SUMMARY Obesity increases the risk for metabolic and cardiovascular diseases and, therefore, may contribute to reduced life expectancy. Adipose tissue is an endocrine organ secreting several hundreds of adipokines, which contribute to the crosstalk of adipose tissue with the brain, liver, muscle and other organs. Altered secretion patterns of adipokines are a symptom of adipose tissue dysfunction and may link obesity to an increased risk of insulin resistance, Type 2 diabetes, fatty liver disease, hypertension, dyslipidemia, endothelial dysfunction, atherosclerosis, dementia, airway disease and some cancers. Adipokines may become clinically important both as biomarkers and as the substrate or target for pharmacotherapeutic management of obesity and its related diseases in the future. This review focuses on selected adipokines, which play a role in glucose homeostasis.

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Obesity significantly increases the risk for metabolic diseases (Type 2 diabetes [T2D], fatty liver disease and dyslipidemia), cardiovascular disorders (hypertension, coronary heart disease and stroke), diseases of the CNS (dementia), obstructive sleep apnea and different types of cancer [1–4]. Adipose tissue may be considered an endocrine gland, which releases bioactive molecules, so called adipokines **(Figure 1)**. Adipokines are peptides or cytokines that are secreted by adipose tissue. To date, more than 600 secretory proteins have been suggested to be secreted from adipose tissue and several of them play an important role in glucose homeostasis [5]. Adipose tissue-derived signals participate in various metabolic processes including the regulation of appetite control, fat distribution, insulin sensitivity and insulin secretion, energy expenditure, and inflammation [4–6]. Adipokines may play a significant role in the regulation of adipogenesis, chemoattraction of immune cells into adipose tissue, adipocyte function via autocrine/paracrine signaling [6]. In addition to the role of adipokines within adipose tissue, they exert important systemic (endocrine) effects, including regulation of appetite, energy expenditure and spontaneous activity, insulin sensitivity and energy metabolism in the brain and peripheral target tissues **(Figure 2)** [4–6]. The adipokine secretion pattern reflects adipose tissue function [4] and may have clinical relevance to evaluate the risk of an individual to develop metabolic and cardiovascular diseases associated with or caused by obesity [1,4–8]. In parallel with or as a consequence of adipose tissue inflammation and dysfunction, secretion of adipose tissue derived factors is significantly altered towards a diabetogenic, proinflammatory and atherogenic adipokine pattern [1,4–8].

The major breakthrough establishing adipose tissue as an endocrine organ was the discovery

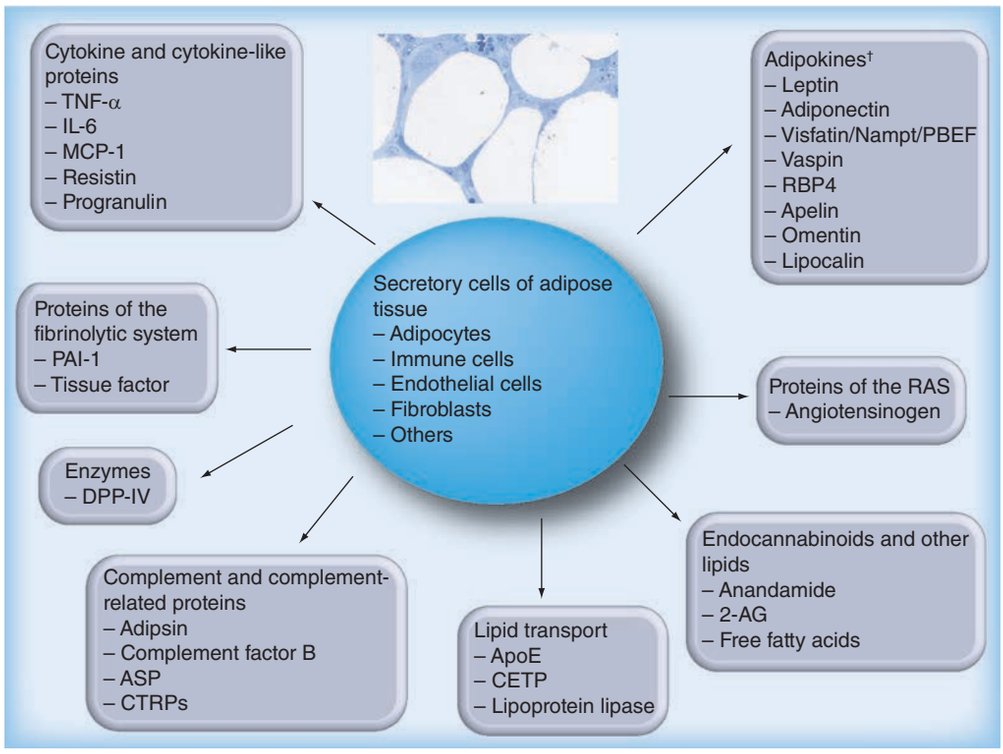

Figure 1. Adipose tissue as an endocrine organ. Several cell types of adipose tissue, including adipocytes, immune cells, fibroblasts and endothelial cells, contribute to adipokine secretion. Examples of endocrine adipose tissue-derived molecules that may contribute to the link between increased adipose tissue and adipose tissue dysfunction in obesity and glucose homeostasis.
†Not otherwise classified.
2-AG: 2-Arachidonoylglycerol; RAS: Renin angiotensin system.
Adapted from [6], data taken from [10].

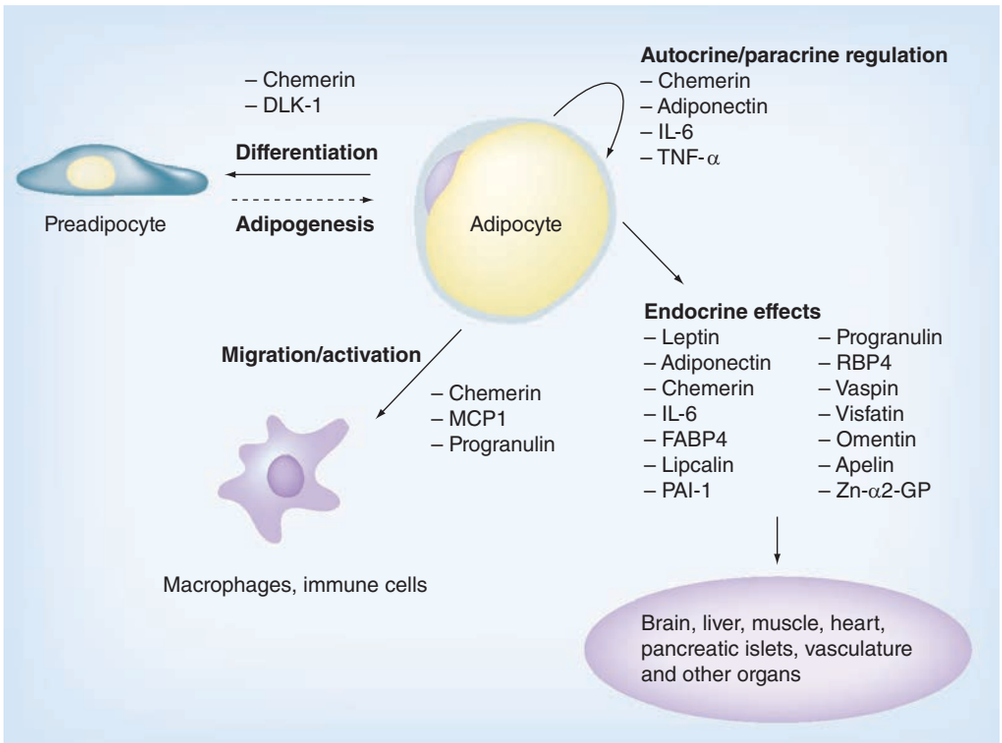

Figure 2. Effects of adipokines on adipogenesis, immune cell migration into adipose tissue, autocrine/paracrine adipocyte regulation and systemic effects. Adipokines have endocrine effects on appetite control, regulation of energy expenditure and activity, influence insulin sensitivity and energy metabolism in insulin sensitive tissues, such as liver, muscle and fat, as well as insulin secretion in pancreatic β-cells.

of leptin as an adipokine, signalling the energy status of the periphery to the CNS [9]. Following leptin, several other adipokines have been discovered that may regulate insulin sensitivity, lipid and glucose metabolism, and endothelial function [4–6]. Adipose tissue releases proinflammatory cytokines/adipokines, interleukins, chemoattractant cytokines, RBP4, serine protease inhibitors, fetuin-A, omentin, bone morphogenetic proteins, clusterin, fractalkine, orosomucoid, FABP4, fibrinogen, haptoglobin, angiopoietin-related proteins, metallothionein, complement factors and SAA proteins among others **(Table 1 & Figure 1)** [4–6,10–12]. The majority of these adipokines are elevated in the obese and correlate with measures of fat mass, fat distribution and insulin sensitivity [6,13]. Interestingly, secretion of factors that are considered anti-inflammatory mediators (e.g., adiponectin, apelin and IL-10) is significantly downregulated in obese states [11,14]. Importantly, the value of adipokine measurements as biomarkers is still limited, since common diseases or pathomechanisms including obesity, insulin resistance and visceral adiposity are rather easy to diagnose and do not necessarily require an adipokine marker. However, there may be a potential use of adipokines as markers of adipose tissue inflammation and function in the future. Adipokines may only be considered as surrogate markers that do not reflect biological processes better than established markers (e.g., CrP for inflammation, high-density lipoprotein [HDL-cholesterol and triglycerides for the metabolic syndrome). However, there is a need to identify biomarkers for the prediction of individual treatment responses to weight loss strategies, to determine diet adherence or dynamics in fat distribution in intervention studies. Research to establish adipokines as such markers has not been very successful in the past. In addition, the value of suggested adipokines in the prediction of diseases needs to be evaluated in epidemiologic studies.

In the context of this overview not all of the >600 adipokines and their potential impact on glucose metabolism can be discussed. In addition, for some of the more recently identified adipokines, the clinical relevance of increased

Table 1. Examples of adipokines and their main function(s).

Adipokine	Function (keywords)
Leptin	Regulation of satiety, appetite, food intake, activity and energy expenditure, fertility, atherogenesis, growth induction
Adiponectin	Antidiabetic, antiatherogenic, anti-inflammatory
Chemerin	Chemoattractant protein, regulation of adipogenesis
TNF-α	Proinflammatory
IL-6	Proinflammatory
PAI-1	Inhibition of plasminogen activators, atherogenesis
FABP-4	Related to Type 2 diabetes risk, myocardial contractility
RBP4	Related to insulin resistance, visceral fat distribution
Vaspin	Serine protease inhibitor, decreases food intake, improves hyperglycemia
Apelin	Inhibits insulin secretion
Nesfatin-1	Direct glucose-dependent insulinotropic effect on β cells
Visfatin/PBEF/Nampt	Nampt-mediated systemic NAD biosynthesis is critical for β-cell function
MCP-1	Chemoattractant protein, adipose tissue inflammation
Progranulin	Chemoattractant protein, neurodegenerative diseases, adipose tissue inflammation
Fetuin-A	Lipid-induced inflammation, insulin resistance, promotes cancer progression
Omentin	Anti-inflammatory
DPP-4	Degrades GIP and GLP-1, inhibitors in clinical use for Type 2 diabetes
Clusterin	Promotes tumor progression and angiogenesis

or decreased plasma concentrations in the context of glucose homeostasis is either not clear or is controversial. Therefore, this review focuses on the subjective selection of classical adipokines including leptin and adiponectin and the more recently identified adipokines RBP4 [15,16], chemerin [17,18], apelin [19,20], vaspin [21–23], DPP-4 [24], visfatin/PBEF/Nampt, nesfatin-1 and cathepsins. The list of adipokines is expanding and very recently more than 40 novel adipokines have been identified by a proteomics approach for primary human adipocytes [25,26].

Mechanisms in which adipokine secretion may affect glucose homeostasis

In principle, there are direct and indirect mechanisms in which altered adipokine secretion may contribute to changes in glucose homeostasis. Direct mechanisms include a role of adipokines in the regulation of insulin secretion and insulin sensitivity at the level of the whole body, organ specific in the liver, brain, muscle and other tissues or on target cells. Indirect mechanisms are mainly related to the fact that adipokines may play a role in inflammation, adipose tissue accumulation and adverse distribution, which subsequently impacts on glucose metabolism. Some adipokines, including adiponectin and CTRP12, have been shown to improve insulin sensitivity [27,28]. Effects on β-cell function have been reported for leptin, which suppresses insulin secretion and adiponectin, which enhances insulin secretion by stimulating both the expression of the insulin gene and exocytosis of insulin granules [29]. Other adipokines have been shown to be associated with abdominal fat distribution (e.g., omentin, RBP4, chemerin, progranulin and vaspin) and adipose tissue dysfunction, including inflammation (e.g., MCP-1, progranulin and chemerin) [5,6].

Leptin

Leptin is secreted from adipocytes and plays an important role in the regulation of satiety, appetite, food intake, activity and energy expenditure, fertility, atherogenesis and growth-inducing properties [29,30]. In the hypothalamus, leptin increases anorexigenic peptide synthesis and decreases orexigenic peptide synthesis, subsequently resulting in reduced appetite [31]. Leptin was discovered in 1994 as the protein product of the *ob* gene [9]. This discovery marked the identification of the mechanism underlying the extremely obese phenotype of the *ob*/*ob* mouse model, which carries a mutation in the *ob* gene, subsequently leading to leptin deficiency [9]. Interestingly, mice that bare a mutation in the leptin receptor gene (*db*/*db* mice) are characterized by obesity and diabetes, and further support the importance of leptin-related mechanisms in the development of obesity [32]. The relationship of leptin with insulin resistance, obesity and cardiovascular disease has been extensively studied since its discovery [29]. Obesity is associated with increased circulating leptin concentrations, which have been suggested to play a role in glucose homeostasis [1,30]. Although exogenous leptin administration significantly increases leptin serum concentrations, it did not alter satiety or appetite, most likely owing to central leptin resistance [33]. However, in extremely rare cases of human genetic leptin deficiency or under conditions of lipodystrophy (a condition that is also associated with low circulating leptin levels), leptin administration dramatically improves insulin sensitivity, glucose and lipid metabolism, and lowers the risk of pancreatitis induced by hypertriglyceridemia [34–35]. The reported insulin sensitizing effects of leptin have been related to increased fatty acid oxidation and decreased triglyceride storage in muscle [36,37]. In the context of leptin's role in

glucose homeostasis, it is important that leptin has been identified as an important regulator of β-cell mass and cell survival [29,38]. β cells express leptin receptors, which mediate direct inhibitory effects of leptin on insulin secretion [38]. Leptin effects on pancreatic β cells may also have significant long-term effects with regards to β-cell proliferation, apoptosis, cell size and inhibition of insulin gene expression [38]. In this context, Zucker diabetic fatty rats, which have a defect in the leptin receptor, are characterized by a reduction in β-cell mass, which is primarily due to an increased rate of β-cell death [39]. Taken together, leptin participates in the regulation of glucose homeostasis at different levels of modulation of the pancreatic β-cell population, which may be aggravated under conditions of hyperleptinemia, such as obesity [38].

Adiponectin

Adiponectin is highly expressed in adipocytes and has been shown to exert important antidiabetic, antiatherogenic and anti-inflammatory properties [27]. Circulation concentrations of adiponectin are lower in patients with obesity, T2D and other insulin-resistant states compared with lean, normal, glucose-tolerant and insulin-sensitive individuals [1,30]. Importantly, it has been shown that adiponectin expression in adipose tissue and secretion increase in parallel with improved insulin sensitivity and weight loss [27]. It has been suggested that insulin-sensitizing effects of glitazones are, at least in part, mediated by their effect on increasing adiponectin serum concentrations [40]. On the other hand, hormones associated with insulin resistance and obesity, including catecholamines, glucocorticoids, TNF-α and other cytokines, have been shown to negatively affect adiponectin expression and secretion *in vitro* [41]. In addition to these effects, adiponectin acts in the brain to increase energy expenditure and cause weight loss [27,40].

The role of adiponectin as a molecule that sensitizes peripheral tissues to insulin has been discovered using models of disrupted adiponectin gene expression in mice [42,43]. Under experimental conditions of adiponectin deficiency using adiponectin knockout mice, impaired insulin sensitivity has been a reproducible finding [42,43]. By contrast, adiponectin may have antiobesity effects by increasing energy expenditure and effects on the differentiation of fat cells in mice with transgenic adiponectin overexpression [44].

The effects of adiponectin on glucose homeostasis may be due to adiponectin's effects on both insulin sensitivity and secretion [29]. Adiponectin could directly improve peripheral insulin sensitivity [27] most likely by reducing glucose levels and increasing fatty acid combustion in myocytes (mediated by the globular C-terminal fragment) [45]. In addition, adiponectin improves insulin sensitivity by autocrine action in fat cells [41] and most likely also in hepatocytes [27]. However, studies on the effects of adiponectin on insulin release revealed inconsistent results [29]. Transgenic *ob/ob* mice with a higher expression of adiponectin's globular domain are characterized by improved insulin sensitivity and enhanced insulin secretion in comparison with control mice [46]. Taken together, there are several lines of evidence that adiponectin may exert insulin-sensitizing effects and protective effects on β-cells [29]. Further *in vivo* studies in rodent model systems clearly showed that systemic recombinant adiponectin treatment results in enhanced insulin secretion [47].

RBP4

RBP4 can be considered a hepato-adipokine, because it is released both from the liver and adipose tissue [15]. In patients with obesity and insulin resistance, elevated RBP4 serum concentrations may be derived from increased RBP4 expression in visceral adipose tissue [48]. The potential association of RBP4 with alterations in glucose metabolism has been identified in mice with an adipose tissue-specific disruption of the GLUT4 transporter [15]. Adipose tissue-selective *Glut4* knockout mice have significantly higher RBP4 serum concentrations compared with control littermates [15]. RBP4 has, therefore, been suggested as a molecule that is important for fuel sensing in adipocytes [1,6,16,30]. The effects of RBP4 are mediated through retinol-dependent and independent mechanisms [6]. RBP4 could induce the gluconeogenic enzyme PEPCK, increase cellular glucose production at the basal state and impair insulin action in hepatocytes *in vitro* [15]. In hepatocytes, reduced RAR-α signaling causes hepatic steatosis, supporting the hypothesis that a defect in retinoic acid signaling could lead to elevated RBP4 serum concentrations that contribute to hepatic steatosis [49]. Administration of recombinantly produced RBP4 to mice impairs-insulin-induced suppression of hepatic glucose production [15]. Experimental upregulation of RBP4 in mice has

been shown to cause insulin resistance through induction of hepatic expression of PEPCK and impairment of skeletal muscle insulin signaling [15]. Moreover, it has been recently demonstrated that RBP4 could lead to insulin resistance by inducing an inflammatory state in adipose tissue via proinflammatory cytokine activation in macrophages [50].

Many studies have been conducted after the notion that serum RBP4 is significantly higher in individuals (as well as mouse models) with insulin resistance [15] to further characterize the role of RBP4 in the development of obesity and T2D. Several of the studies support the observation that increased RBP4 serum concentrations are associated with obesity, insulin resistance and visceral fat accumulation [16,48]. Therefore, increased RBP4 serum concentrations may represent an important connection between visceral obesity and impaired insulin sensitivity and several metabolic diseases [8]. RBP4 does not seem to play a direct role in the regulation of insulin secretion. We recently demonstrated that circulating RBP4 reflects weight loss and weight regain in response to different diets. These data may suggest that RBP4 only reflects body weight dynamics and subsequent changes in glucose homeostasis [13].

Chemerin

Chemerin (RARRES2 or TIG2) is a molecule secreted from adipose tissue and the liver with chemoattractant properties. It has been demonstrated that chemerin plays a role in the regulation of adipogenesis and may induce insulin resistance [17]. In patients with obesity and impaired insulin sensitivity, but also with chronic inflammation, chemerin serum concentrations are elevated [17,18]. Chemerin is released as an 18-kDa inactive proprotein, which undergoes cleavage by an extracellular protease generating the 16-kDa active chemerin [17,51]. On activated macrophages, which express either the CMKLR1 or the ChemR23 receptor, chemerin may have important anti-inflammatory effects [51,52]. Chemerin contributes to the regulation of adipogenesis, differentiation and proliferation of fat cells, and modulates the expression of genes involved in glucose and lipid homeostasis [1,6,30]. Chemerin enhances insulin-stimulated glucose uptake *in vitro*, suggesting that chemerin may increase insulin sensitivity in adipose tissue [53]. Mice with a disruption of the ChemR23 receptor are characterized by an increased neutrophil infiltrate following lipopolysaccharide challenge due to a diminished chemerin response [54]. A similar phenotype has been observed in mice with acute lipopolysaccharide-induced lung inflammation [54]. Here, chemerin was also shown to have anti-inflammatory properties, reducing neutrophil infiltration and proinflammatory cytokine release in a ChemR23-dependent manner [54].

A mechanistic role of chemerin in the development of impaired glucose metabolism has not been established yet. However, in individuals with insulin-sensitive obesity, chemerin serum concentrations are significantly higher than those of age-, gender- and BMI-matched controls [55]. Moreover, circulating chemerin levels are associated with bodyweight and serum triglyceride concentrations, as well as blood pressure in normal subjects [56,57]. In explants from the adipose tissue of patients with obesity, significantly higher chemerin secretion has been found compared with lean controls [58]. These studies further suggested that chemerin release correlates with BMI, WHR, adipocyte size and insulin resistance in adipocytes [58]. Sell *et al.* suggested that secretion of chemerin from adipocytes may be involved in the negative crosstalk between adipose tissue and skeletal muscle, contributing to insulin resistance [58]. In another recent study, increased circulating and adipose tissue chemerin levels in women with polycystic ovary syndrome were reported to be upregulated by insulin, whereas 6 months of metformin treatment significantly decreased serum chemerin in these women [56]. A comparison of chemerin levels circulating either in portal, hepatic or systemic venous blood demonstrated that elevated systemic chemerin concentrations in obesity and its related metabolic diseases seem to be associated with a proinflammatory state rather than BMI or abdominal visceral adipose tissue mass [57]. We recently postulated that chemerin may initiate adipose tissue dysfunction and suggested that reduced expression of chemerin in adipose tissue may contribute to improved insulin sensitivity and subclinical inflammation beyond significant weight loss [18]. However, further mechanistic experiments and clinical studies are required to define whether chemerin contributes to the altered communication between adipose tissue and other organs in obesity.

Apelin

The adipokine apelin has been suggested to contribute to the regulation of glucose

metabolism [19,20]. Apelin is a 36-amino acid peptide and the endogenous ligand of the G-protein-coupled APJ receptor. Apelin is expressed in the CNS (particularly high expression in the hypothalamus), heart, skeletal muscle, gastrointestinal organs (e.g., stomach) and adipose tissue [59,60]. There are several active apelin forms including apelin-36, apelin-17, apelin-13 and the pyroglutamated form of apelin-13 [20,59,60]. Apelin and *APJ* mRNA are widely expressed in several rat and human tissues and play a role in the regulation of cardiovascular homeostasis, food intake, cell proliferation and angiogenesis [59,60].

Mice with a disruption of the apelin gene do not exhibit significant metabolic alterations [61]. However, aging apelin knockout mice progressively develop a dysfunction of cardiac contractility, which is associated with systolic dysfunction [61]. These experiments support the notion that apelin may play a major role in maintenance of cardiac contractility in pressure overload and aging [61]. Hypoxia plays a role in the development of adipose tissue dysfunction. In this context, it has been suggested that apelin expression may be induced by hypoxia, both in endothelial and vascular smooth muscle cells mediated by the binding of HIF-1 to a HRE located within the first intron of the apelin gene [62].

Considering a potential role of apelin in the regulation of glucose homeostasis, there is clinical evidence for elevated circulating apelin levels in obese and insulin-resistant individuals [19]. In addition, an association between circulating apelin and liver cirrhosis has been reported both in rats and humans [20]. In line with these observational data, application of an apelin receptor antagonist to rats with liver cirrhosis caused diminished hepatic fibrosis and a loss of ascites, suggesting the hepatic apelin system as a novel therapeutic target in liver disease [63].

Apelin inhibits insulin secretion in mice [64], suggesting a link between apelin and glucose homeostasis. Circulating apelin correlates with hyperinsulinemia and bodyweight, suggesting that apelin may link adipose tissue dysfunction to obesity-related diseases [8,19]. In this context, apelin seems to have effects associated with enhanced glucose utilization in skeletal muscle and fat, which subsequently contribute to lower circulating glucose concentrations [65]. Moreover, experimental apelin administration improved glucose tolerance and glucose utilization in mouse models of diet-induced obesity and insulin resistance [65]. These beneficial effects of apelin on glucose metabolism involve activation of endothelial nitric oxide synthase, AMP-activated protein kinase and Akt. Collectively, human and rodent data suggest that apelin contributes to the regulation of glucose homeostasis and may link increased adipose tissue mass to obesity-related metabolic and inflammatory diseases.

Visceral adipose tissue-derived serpin

Visceral adipose tissue-derived serpin (vaspin) was identified as a member of the serine protease inhibitor family. Vaspin has been found to be highly expressed in visceral adipose tissue of Otsuka Long-Evans Tokushima Fatty rats at the age when obesity and insulin plasma concentrations reach a peak [21]. Although vaspin is clearly expressed in human adipose tissue [66], the name vaspin may be a little misleading since we demonstrated expression of vaspin in skin, stomach and rodent hypothalamus and pancreatic islets [23,67]. So far, the mechanisms of how vaspin secretion may be linked to impaired glucose metabolism and insulin sensitivity are not entirely understood. We recently demonstrated that vaspin inhibits one of its target proteases, kallikrein 7, which is able to cleave insulin at least *in vitro* [68]. Improved glucose metabolism in response to vaspin treatment could, therefore, be mediated by increased insulin plasma concentrations due to the inhibition of insulin degradation in the circulation by kallikrein 7 [68]. In Otsuka Long-Evans Tokushima Fatty rats, vaspin protein and mRNA expression, as well as vaspin serum concentrations increase with worsening of diabetes and bodyweight loss at 50 weeks and could be normalized with insulin or pioglitazone treatment [21]. Vaspin administration to mouse models of obesity improves insulin sensitivity, glucose tolerance and changed mRNA expression of insulin resistance candidate genes [21]. We recently provided experimental evidence that treatment of different mouse models with recombinant vaspin leads to sustained glucose lowering and reduction of food intake [67]. Significantly, adipose tissue vaspin mRNA is almost exclusively found in adipose tissue of obese, but not lean, individuals, with significant differences between higher visceral and low subcutaneous expression [66]. Elevated vaspin serum concentrations are associated with obesity, impaired insulin sensitivity and fitness level and leptin serum concentrations [22]. In contrast with previous data [22,66], a comparison between BMI-,

age, and gender-matched insulin-sensitive versus insulin-resistant obese individuals revealed indistinguishable vaspin levels between these groups [55]. These somewhat contradictory data suggest that the association between circulating vaspin, fat distribution and insulin sensitivity is more complex than previously thought. However, in the future, vaspin serum concentrations could be used as a biomarker to monitor the individual success of weight loss interventions. In this context, we found that vaspin closely reflects a healthier dietary pattern by analyzing data from 322 participants of the 2-year DIRECT [13,69].

DPP-4

Intake of nutrients may stimulate the release of the incretin hormones, including GIP and GLP-1 [70,71]. Incretins have been shown to enhance glucose-dependent insulin secretion and inhibit glucagon secretion, which together may reduce hepatic gluconeogenesis [71]. The effect of incretins is significantly reduced in patients with T2D and contributes to impaired insulin secretion and chronic hyperglycemia [71]. Under normal physiological conditions, DPP-4 rapidly degrades GIP and GLP-1 [72]. Inhibition of DPP-4 is a therapeutic option to lower hyperglycemia in patients with T2D. DPP-4 inhibitors are a relatively new class of oral glucose-lowering drugs. DPP-4 inhibitors exert their hypoglycemic effect indirectly by increasing plasma concentration, duration and action of incretins [71,73]. Currently, the DPP-4 inhibitors alogliptin, sitagliptin, linagliptin, saxagliptin and vildagliptin are in clinical use as antidiabetic drugs to improve glycemic control by stimulating glucose-induced pancreatic insulin secretion and suppressing glucagon production [74]. Although research has focused on the role of DPP-4 in the degradation of GLP-1, recent data suggest that DPP-4 also exerts direct effects, as it is able to induce insulin resistance in adipocytes and skeletal muscle cells in concentrations that can be found in the circulation of overweight and obese subjects [24]. This notion is also supported by the notion that DPP-4 inhibitors might also exert incretin-independent effects, further suggesting that DPP-4 might have direct metabolic effects [73]. The fact that circulating DPP-4 and DPP-4 expression in adipose tissue correlate with adipocyte size and adipose tissue inflammation [24] might also suggest that proinflammatory adipokines released from enlarged adipocytes could regulate DPP-4 release. Therefore, DPP-4 might not only reflect adipose tissue function, but it may also have local effects within adipose tissue and systemic effects. Lamers *et al.* recently demonstrated that adipocytes release DPP-4 in a differentiation-dependent manner [24]. DPP-4 is a 766-amino acid, membrane-associated, serine protease enzyme [72]. The enzyme is widely detected in numerous tissues, such as kidney, liver, intestine, spleen, lymphocytic organs, placenta, adrenal glands and vascular endothelium [73]. Using proteomic profiling of the human adipocyte secretome, DPP-4 has been identified as a novel adipokine [24]. Higher secretion of DPP-4 from enlarged adipose tissue could impair insulin sensitivity at the adipose organ level in an autocrine/paracrine mode [24]. Lamers *et al.* recently provided evidence that DPP-4 release correlates with fat cell size [24], suggesting that DPP-4 may, as well as several other adipokines, link adipose tissue dysfunction and adipocyte hypertrophy to impaired whole-body glucose metabolism. Increased DPP-4 activity and serum concentrations in obesity may serve as a model of how altered adipokine secretion may be successfully used as a therapeutic target in the treatment of obesity-related diseases. However, further work is needed to elucidate the functional role of DPP-4 within adipose tissue and to define whether higher DPP-4 expression and serum concentration may contribute to higher efficacy of DPP-4 inhibitors in patients with obesity-associated T2D.

Visfatin/PBEF/Nampt

The *PBEF1* gene encodes a polypeptide of 491 amino acids with a molecular mass of 52 kDa that has been isolated from a human peripheral blood lymphocyte cDNA library [75]. It has first been described as an adipokine predominantly secreted from visceral fat exerting insulin-mimetic effects [76]; however, subsequent human studies revealed that other tissues and adipose tissue depots may also express visfatin/PBEF/Nampt and the effects of this molecule as an insulin mimetic are controversial [75,77]. Visfatin/PBEF/Nampt has an important enzymatic function in synthesizing NMN from nicotinamide and PRPP [75]. Recently, it has been demonstrated that hypercaloric feeding, as well as aging, compromise NAMPT-mediated NAD^+ biosynthesis and may, therefore, contribute to the pathogenesis of T2D [78]. Yoshino *et al.* recently demonstrated that administration of NMN to mouse models of obesity and

T2D promotes NAD^+ biosynthesis, thereby ameliorating glucose intolerance and improving hepatic insulin sensitivity [78]. The mechanism by which visfatin/PBEF/Nampt contributes to alterations in glucose homeostasis may involve regulation of expression of genes related to oxidative stress, inflammatory response and circadian rhythm, at least in part via SIRT1 activation [78]. Revollo *et al.* demonstrated that Nampt-mediated systemic NAD biosynthesis is critical for β-cell function [79], whereas circulating visfatin/PBEF/Nampt (in contradiction to the initial observations [76]) does not exert insulin-mimetic effects *in vitro* or *in vivo*. Taken together, the role of visfatin/PBEF/Nampt in glucose homeostasis may be related to recent evidence that Nampt-mediated systemic NAD biosynthesis is a critical contributor to β-cell function [78].

Nesfatin-1

Nesfatin-1 is a 82-amino acid peptide derived from the precursor protein, NUCB2, which has been proposed as a novel satiety molecule acting through leptin-independent mechanisms [80]. Nesfatin-1 is expressed in the CNS, the pituitary gland, stomach, pancreas, testis and also adipose tissue [80]. It has been shown that nesfatin-1 stimulates the pituitary–adrenal axis and sympathetic nervous system, and influences visceral functions and emotion [80]. Since activation of NUCB2/nesfatin-1 neurons in the brain is mediated by different stressors, it has been suggested that nesfatin-1 plays a role in the adaptive response under stressful conditions [80]. In addition to these central effects, there is accumulating evidence that nesfatin-1 exerts direct glucose-dependent insulinotropic action and increases preproinsulin mRNA expression in rodent-isolated islets or cultured mouse insulinoma cell line cells [81,82]. Importantly, intravenous but not central administration of recombinant nesfatin-1 to *db/db* mice significantly reduces hyperglycemia, further supporting the hypothesis that nesfatin-1 has a direct glucose-dependent insulinotropic effect on β cells [83]. Further studies are necessary, including the identification of the nesfatin-1 receptor(s) to improve our understanding of nesfatin-1 cellular signal transduction and related mechanisms.

Cathepsins

Cathepsins belong to a family of cysteine proteases and are an important class of enzymes released from adipose tissue [84]. Cathepsins S, L and K may play a role in the development of atherosclerosis, since they are abundantly expressed in atherosclerotic lesions in humans [85]. Among the cathepsins, cathepsin S has been shown to be increasingly expressed in adipose tissue with the development of obesity and both adipose tissue mRNA expression and serum concentrations correlate with measures of obesity [86]. Adipose tissue also expresses cathepsin K [87] and L [88], and these cathepsins have been proposed to contribute to the link between altered adipose tissue function, endothelial dysfunction and atherosclerosis [89]. Cathepsin may also play an important role in the regulation of whole-body glucose metabolism. Mice with a disruption of cathepsin K or L have reduced adipose tissue mass and show evidence for improved glucose metabolism [88,90]. Interestingly, deletion of these cathepsins caused a beneficial metabolic phenotype despite high fat diet challenges, suggesting that pharmacological inhibition of (these) cathepsins may target both the increased risk of developing T2D and premature atherosclerosis in obese individuals. Further studies are required to address the question of which member of the cathepsin family is most clinically relevant in obese states. Naour *et al.* recently found that in human obesity cathepsin S is more strongly affected by changes in energy balance compared with cathepsins L and K, both in adipose tissue and systemically [89]. The mechanism of cathepsin action in linking obesity to its related metabolic and cardiovascular diseases may involve the regulation of adipose tissue derived hormones. Supporting this hypothesis, it has be show that cathepsin S processes leptin and inactivates its biological activity.

Summary & conclusion

Adipose tissue produces many (most likely more than 600) bioactive molecules, called adipokines. Changes in adipokine secretion may link impaired adipose tissue function in obesity to obesity-related disorders, including T2D. Adipokines are important modulators of glucose metabolism, because they may primarily contribute to adverse fat distribution (chemerin, RBP4 and others), altered appetite and satiety (e.g., leptin, vaspin and nesfatin-1), impaired insulin sensitivity (adiponectin, leptin, RBP4 and others) or insulin secretion (e.g., DPP-4 and nesfatin-1), and to

inflammation (e.g., resistin, IL-6, TNF-α, chemerin and progranulin).

In addition, adipokines are important as biomarkers and have the potential for future pharmacological treatment strategies for obesity and its related diseases.

Future perspective

Functional characterization of newly identified adipokines is a major unmet need in future obesity research. The identification of mechanisms of how adipokines may affect insulin sensitivity, glucose metabolism and endothelial function could be the basis for novel pharmacological treatment approaches of obesity and its related disorders.

Financial & competing interests disclosure

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References

Papers of special note have been highlighted as:

- ▪ of interest
- ▪▪ of considerable interest

1 Van Gaal LF, Mertens IL, De Block CE. Mechanisms linking obesity with cardiovascular disease. *Nature* 444, 875–880 (2006).

▪ **Very good overview of current concepts of how obesity may be linked to cardiovascular disease.**

2 LeRoith D, Novosyadlyy R, Gallagher EJ *et al.* Obesity and Type 2 diabetes are associated with an increased risk of developing cancer and a worse prognosis; epidemiological and mechanistic evidence. *Exp. Clin. Endocrinol. Diabetes* 116, S4–S6 (2008).

3 Kahn SE, Hull RL, Utzschneider KM. Mechanisms linking obesity to insulin resistance and Type 2 diabetes. *Nature* 444, 840–846 (2006).

4 Blüher M. Adipose tissue dysfunction in obesity. *Exp. Clin. Endocrinol. Diabetes* 117, 241–250 (2009).

5 Lehr S, Hartwig S, Sell H. Adipokines: a treasure trove for the discovery of biomarkers for metabolic disorders. *Proteomics Clin. Appl.* 6(1–2), 91–101 (2012).

6 Blüher M. Do adipokines link obesity to its related metabolic and cardiovascular diseases? *Clin. Lipidol.* 5, 95–107 (2010).

7 Bays HE. 'Sick fat,' metabolic disease, and atherosclerosis. *Am. J. Med.* 122, S26–S37 (2009).

8 Blüher M. Clinical relevance of adipokines. *Diabetes Metab. J.* 36, 317–327 (2012).

9 Zhang Y, Proenca R, Maffei M *et al.* Positional cloning of the mouse obese gene and its human homologue. *Nature* 372, 425–432 (1994).

▪▪ **Describes the discovery of leptin.**

10 Kershaw EE, Flier JS. Adipose tissue as an endocrine organ. *J. Clin. Endocrinol. Metab.* 89, 2548–2556 (2004).

11 Schinner S, Kempf K, Overmann H *et al.* Association of impaired glucose metabolism in morbid obesity with hypoadiponectinaemia. *Exp. Clin. Endocrinol. Diabetes* 116, S64–S69 (2008).

12 Wajchenberg BL. Subcutaneous and visceral adipose tissue: their relation to the metabolic syndrome. *Endocr. Rev.* 21, 697–738 (2000).

▪▪ **Comprehensive overview on the differences between visceral and subcutaneous adipose tissue.**

13 Blüher M, Rudich A, Klöting N *et al.* Two patterns of adipokine and other biomarker dynamics in a long-term weight loss intervention. *Diabetes Care* 35, 342–349 (2012).

14 Stefan N, Stumvoll M. Adiponectin – its role in metabolism and beyond. *Horm. Metab. Res.* 34, 469–474 (2002).

15 Yang Q, Graham TE, Mody N *et al.* Serum retinol binding protein 4 contributes to insulin resistance in obesity and Type 2 diabetes. *Nature* 436, 356–362 (2005).

▪▪ **Seminal study about the role of RBP4 in metabolic diseases.**

16 Graham TE, Yang Q, Blüher M *et al.* Retinol-binding protein 4 and insulin resistance in lean, obese, and diabetic subjects. *N. Engl. J. Med.* 354, 2552–2563 (2006).

17 Wittamer V, Franssen JD, Vulcano M *et al.* Specific recruitment of antigen-presenting cells by chemerin, a novel processed ligand from human inflammatory fluids. *J. Exp. Med.* 198, 977–985 (2003).

18 Chakaroun R, Raschpichler M, Klöting N *et al.* Effects of weight loss and exercise on chemerin serum concentrations and adipose tissue expression in human obesity. *Metabolism* 61, 706–714 (2012).

19 Boucher J, Masri B, Daviaud D *et al.* Apelin, a newly identified adipokine up-regulated by insulin and obesity. *Endocrinology* 146, 1764–1771 (2005).

20 Castan-Laurell I, Dray C, Attané C *et al.* Apelin, diabetes, and obesity. *Endocrine* 40, 1–9 (2011).

21 Hida K, Wada J, Eguchi J *et al.* Visceral adipose tissue-derived serine protease inhibitor: a unique insulin-sensitizing adipocytokine in obesity. *Proc. Natl Acad. Sci. USA* 102, 10610–10615 (2005).

22 Youn BS, Klöting N, Kratzsch J *et al.* Serum vaspin concentrations in human obesity and Type 2 diabetes. *Diabetes* 57, 372–377 (2008).

23 Blüher M. Vaspin in obesity and diabetes: pathophysiological and clinical significance. *Endocrine* 41, 176–182 (2012).

24 Lamers D, Famulla S, Wronkowitz N *et al.* Dipeptidyl peptidase 4 is a novel adipokine potentially linking obesity to the metabolic syndrome. *Diabetes* 60, 1917–1925 (2011).

25 Lehr S, Hartwig S, Lamers D *et al.* Identification and validation of novel adipokines released from primary human adipocytes. *Mol. Cell. Proteomics* 11, M111.010504 (2012).

26 Dahlman I, Elsen M, Tennagels N *et al.* Functional annotation of the human fat cell secretome. *Arch. Physiol. Biochem.* 118, 84–91 (2012).

27 Matsuzawa Y. The metabolic syndrome and adipocytokines. *FEBS Lett.* 580, 2917–2921 (2006).

28 Wei Z, Peterson JM, Lei X *et al.* C1q/TNF-related protein-12 (CTRP12), a novel adipokine that improves insulin sensitivity and glycemic control in mouse models of obesity and diabetes. *J. Biol. Chem.* 287, 10301–10315 (2012).

29 Lee YH, Magkos F, Mantzoros CS *et al.* Effects of leptin and adiponectin on pancreatic β-cell function. *Metabolism* 60, 1664–1672 (2011).

30 Ernst MC, Sinal CJ. Chemerin: at the crossroads of inflammation and obesity. *Trends Endocrinol. Metab.* 21, 660–667 (2010).

31 Ahima RS, Flier JS. Leptin. *Annu. Rev. Physiol.* 62, 413–437 (2000).

32 Chen H, Charlat O, Tartaglia LA *et al.* Evidence that the diabetes gene encodes the leptin receptor: identification of a mutation in the leptin receptor gene in *db/db* mice. *Cell* 84, 491–495 (1996).

■■ **Identification of the genetic defect in the leptin receptor of diabetic *db/db* mice.**

33 Savage DB, O'Rahilly S. Leptin: a novel therapeutic role in lipodystrophy. *J. Clin. Invest.* 109, 1285–1286 (2002).

34 Oral EA, Simha V, Ruiz E *et al.* Leptin-replacement therapy for lipodystrophy. *N. Engl. J. Med.* 346, 570–578 (2002).

35 Farooqi IS, Jebb SA, Langmack G *et al.* Effects of recombinant leptin therapy in a child with congenital leptin deficiency. *N. Engl. J. Med.* 341, 879–884 (1999).

36 Dyck DJ. Adipokines as regulators of muscle metabolism and insulin sensitivity. *Appl. Physiol. Nutr. Metab.* 34, 396–402 (2009).

37 Yaspelkis BB 3rd, Singh MK, Krisan AD *et al.* Chronic leptin treatment enhances insulin-stimulated glucose disposal in skeletal muscle of high-fat fed rodents. *Life Sci.* 74, 1801–1816 (2004).

38 Marroquí L, Gonzalez A, Ñeco P *et al.* Role of leptin in the pancreatic β-cell: effects and signaling pathways. *J. Mol. Endocrinol.* 49, R9–R17 (2012).

39 Pick A, Clark J, Kubstrup C *et al.* Role of apoptosis in failure of beta-cell mass compensation for insulin resistance and beta-cell defects in the male Zucker diabetic fatty rat. *Diabetes* 47, 358–364 (1998).

40 Kralisch S, Blüher M, Paschke R *et al.* Adipokines and adipocyte targets in the future management of obesity and the metabolic syndrome. *Mini Rev. Med. Chem.* 7, 39–45 (2007).

41 Dietze-Schroeder D, Sell H, Uhlig M, Koenen M, Eckel J. Autocrine action of adiponectin on human fat cells prevents the release of insulin resistance-inducing factors. *Diabetes* 54, 2003–2011 (2005).

42 Kubota N, Terauchi Y, Yamauchi T *et al.* Disruption of adiponectin causes insulin resistance and neointimal formation. *J. Biol. Chem.* 277, 25863–25866 (2002).

■ **Experimental evidence from adiponectin knockout mice for the role of adiponectin in glucose homeostasis.**

43 Maeda N, Shimomura I, Kishida K *et al.* Diet-induced insulin resistance in mice lacking adiponectin/ACRP30. *Nat. Med.* 8, 731–737 (2002).

■ **Experimental evidence from adiponectin knockout mice for the role of adiponectin in glucose homeostasis.**

44 Bauche IB, El Mkadem SA, Pottier AM *et al.* Overexpression of adiponectin targeted to adipose tissue in transgenic mice: impaired adipocyte differentiation. *Endocrinology* 148, 1539–1549 (2007).

45 Fruebis J, Tsao TS, Javorschi S *et al.* Proteolytic cleavage product of 30-kDa adipocyte complement-related protein increases fatty acid oxidation in muscle and causes weight loss in mice. *Proc. Natl Acad. Sci. USA* 98, 2005–2010 (2001).

46 Yamauchi T, Kamon J, Waki H *et al.* Globular adiponectin protected *ob/ob* mice from diabetes and ApoE-deficient mice from atherosclerosis. *J. Biol. Chem.* 278, 2461–2468 (2003).

47 Okamoto M, Ohara-Imaizumi M, Kubota N *et al.* Adiponectin induces insulin secretion *in vitro* and *in vivo* at a low glucose concentration. *Diabetologia* 51, 827–835 (2008).

48 Klöting N, Graham TE, Berndt J *et al.* Serum retinol-binding protein is more highly expressed in visceral than in subcutaneous adipose tissue and is a marker of intra-abdominal fat mass. *Cell Metab.* 6, 79–87 (2007).

49 Shiota G. Loss of function of retinoic acid in liver leads to steatohepatitis and liver tumor: a NASH animal model. *Hepatol. Res.* 33, 155–160 (2005).

50 Norseen J, Hosooka T, Hammarstedt A *et al.* Retinol-binding protein 4 inhibits insulin signaling in adipocytes by inducing proinflammatory cytokines in macrophages through a c-Jun N-terminal kinase- and Toll-like receptor 4-dependent and retinol-independent mechanism. *Mol. Cell. Biol.* 32, 2010–2019 (2012).

51 Bozaoglu K, Bolton K, McMillan J *et al.* Chemerin is a novel adipokine associated with obesity and metabolic syndrome. *Endocrinology* 148, 4687–4694 (2007).

52 Cash JL, Hart R, Russ A *et al.* Synthetic chemerin-derived peptides suppress inflammation through ChemR23. *J. Exp. Med.* 205, 767–775 (2008).

53 Takahashi M, Takahashi Y, Takahashi K *et al.* Chemerin enhances insulin signaling and potentiates insulin-stimulated glucose uptake in 3T3-L1 adipocytes. *FEBS Lett.* 582, 573–578 (2008).

54 Luangsay S, Wittamer V, Bondue B *et al.* Mouse ChemR23 is expressed in dendritic cell subsets and macrophages, and mediates an anti-inflammatory activity of chemerin in a lung disease model. *J. Immunol.* 183, 6489–6499 (2009).

55 Klöting N, Fasshauer M, Dietrich A *et al.* Insulin-sensitive obesity. *Am. J. Physiol. Endocrinol. Metab.* 299, e506–e515 (2010).

56 Tan BK, Chen J, Farhatullah S *et al.* Insulin and metformin regulate circulating and adipose tissue chemerin. *Diabetes* 58, 1971–1977 (2009).

■ **Important clinical study on the regulation of chemerin serum concentration.**

57 Weigert J, Neumeier M, Wanninger J *et al.* Systemic chemerin is related to inflammation rather than obesity in Type 2 diabetes. *Clin. Endocrinol. (Oxf.)* 72, 342–348 (2010).

58 Sell H, Laurencikiene J, Taube A *et al.* Chemerin is a novel adipocyte-derived factor inducing insulin resistance in primary human skeletal muscle cells. *Diabetes* 58, 2731–2740 (2009).

59 Lee DK, Cheng R, Nguyen T *et al.* Characterization of apelin, the ligand for the APJ receptor. *J. Neurochem.* 74, 34–41 (2000).

60 Tatemoto K, Hosoya M, Habata Y *et al.* Isolation and characterization of a novel endogenous peptide ligand for the human APJ receptor. *Biochem. Biophys. Res. Commun.* 251, 471–476 (1998).

61 Kuba K, Zhang L, Imai Y *et al.* Impaired heart contractility in apelin gene-deficient mice associated with aging and pressure overload. *Circ. Res.* 101, e32–e42 (2007).

62 Eyries M, Siegfried G, Ciumas M *et al.* Hypoxia-induced apelin expression regulates endothelial cell proliferation and regenerative angiogenesis. *Circ. Res.* 103, 432–440 (2008).

63 Principe A, Melgar-Lesmes P, Fernández-Varo G *et al.* The hepatic apelin system: a new therapeutic target for liver disease. *Hepatology* 48, 1193–1201 (2008).

64 Sorhede Winzell M, Magnusson C, Ahren B. The apj receptor is expressed in pancreatic islets and its ligand, apelin, inhibits insulin secretion in mice. *Regul. Pept.* 131, 12–17 (2005).

65 Dray C, Knauf C, Daviaud D *et al.* Apelin stimulates glucose utilization in normal and obese insulin-resistant mice. *Cell Metab.* 8, 437–445 (2008).

66 Klöting N, Berndt J, Kralisch S *et al.* Vaspin gene expression in human adipose tissue: association with obesity and Type 2 diabetes. *Biochem. Biophys. Res. Commun.* 339, 430–436 (2006).

67 Klöting N, Kovacs P, Kern M *et al.* Central vaspin administration acutely reduces food intake and has sustained blood glucose-lowering effects. *Diabetologia* 54, 1819–1823 (2011).

68 Heiker JT, Klöting N, Kovacs P *et al.* Vaspin inhibits kallikrein 7 by serpin mechanism. *Cell. Mol. Life Sci.* 70(14), 2569–2583 (2013).

69 Shai I, Schwarzfuchs D, Henkin Y *et al.* Dietary Intervention Randomized Controlled Trial (DIRECT) Group. Weight loss with a low-carbohydrate, Mediterranean, or low-fat diet. *N. Engl. J. Med.* 359, 229–241 (2008).

70 Drucker DJ, Nauck MA. The incretin system: glucagon-like peptide-1 receptor agonists and dipeptidyl peptidase-4 inhibitors in Type 2 diabetes. *Lancet* 368, 1696–1705 (2006).

71 Nauck MA. Incretin-based therapies for Type 2 diabetes mellitus: properties, functions, and clinical implications. *Am. J. Med.* 124, S3–S18 (2011).

72 Holst JJ, Gromada J. Role of incretin hormones in the regulation of insulin secretion in diabetic and nondiabetic humans. *Am. J. Physiol. Endocrinol. Metab.* 287, e199–e206 (2004).

73 Lotfy M, Singh J, Kalász H *et al.* Medicinal chemistry and applications of incretins and DPP-4 inhibitors in the treatment of Type 2 diabetes mellitus. *Open Med. Chem. J.* 5, 82–92 (2011).

74 Deacon CF, Carr RD, Holst JJ. DPP-4 inhibitor therapy: new directions in the treatment of Type 2 diabetes. *Front. Biosci.* 13, 1780–1794 (2008).

75 Sommer G, Garten A, Petzold S *et al.* Visfatin/PBEF/Nampt: structure, regulation and potential function of a novel adipokine. *Clin. Sci. (Lond.)* 115, 13–23 (2008).

76 Fukuhara A, Matsuda M, Nishizawa M *et al.* Visfatin: a protein secreted by visceral fat that mimics the effects of insulin. *Science* 307, 426–430 (2005).

77 Berndt J, Klöting N, Kralisch S. Plasma visfatin concentrations and fat depot-specific mRNA expression in humans. *Diabetes* 54, 2911–2916. (2005).

78 Yoshino J, Mills KF, Yoon MJ, Imai S. Nicotinamide mononucleotide, a key NAD^+ intermediate, treats the pathophysiology of diet- and age-induced diabetes in mice. *Cell Metab.* 14, 528–536 (2011).

79 Revollo JR, Körner A, Mills KF *et al.* Nampt/PBEF/visfatin regulates insulin secretion in beta cells as a systemic NAD biosynthetic enzyme. *Cell Metab.* 6, 363–375 (2007).

80 Stengel A, Taché Y. Minireview: Nesfatin-1 – an emerging new player in the brain-gut, endocrine, and metabolic axis. *Endocrinology* 152, 4033–4038 (2011).

81 Gonzalez R, Reingold BK, Gao X *et al.* Nesfatin-1 exerts a direct, glucose-dependent insulinotropic action on mouse islet β- and MIN6 cells. *J. Endocrinol.* 208, R9–R16 (2011).

82 Nakata M, Manaka K, Yamamoto S, Mori M, Yada T. Nesfatin-1 enhances glucose-induced insulin secretion by promoting Ca^{2+} influx through L-type channels in mouse islet β-cells. *Endocr. J.* 58, 305–313 (2011).

83 Su Y, Zhang J, Tang Y, Bi F, Liu JN. The novel function of nesfatin-1: anti-hyperglycemia. *Biochem. Biophys. Res. Commun.* 391, 1039–1042 (2010).

84 Lecaille F, Kaleta J, Brömme D. Human and parasitic papain-like cysteine proteases: their role in physiology and pathology and recent developments in inhibitor design. *Chem. Rev.* 102, 4459–4488 (2002).

85 Sukhova GK, Shi GP, Simon DI, Chapman HA, Libby P. Expression of the elastolytic cathepsins S and K in human atheroma and regulation of their production in smooth muscle cells. *J. Clin. Invest.* 102, 576–583 (1998).

86 Taleb S, Lacasa D, Bastard JP *et al.* Cathepsin S, a novel biomarker of adiposity: relevance to atherogenesis. *FASEB J.* 19, 1540–1542 (2005).

87 Chiellini C, Costa M, Novelli SE *et al.* Identification of cathepsin K as a novel marker of adiposity in white adipose tissue. *J. Cell. Physiol.* 195, 309–321 (2003).

88 Yang M, Zhang Y, Pan J. Cathepsin L activity controls adipogenesis and glucose tolerance. *Nat. Cell. Biol.* 9, 970–977 (2007).

89 Naour N, Rouault C, Fellahi S. Cathepsins in human obesity: changes in energy balance predominantly affect cathepsin S in adipose tissue and in circulation. *J. Clin. Endocrinol. Metab.* 95, 1861–1868 (2010).

90 Yang M, Sun J, Zhang T. Deficiency and inhibition of cathepsin K reduce body weight gain and increase glucose metabolism in mice. *Arterioscler. Thromb. Vasc. Biol.* 28, 2202–2208 (2008).