

Adipocyte dysfunction, inflammation and metabolic syndrome

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Abstract Obesity is frequently associated with chronic inflammation, metabolic and vascular alterations which predispose to the development of the Metabolic Syndrome (MetS). However, the individual obesity-related risk for the MetS is not determined by increased fat mass alone. Heterogeneity of body composition, fat distribution and adipose tissue (AT) function may underly the variable risk to develop metabolic and cardiovascular diseases associated with increased body fat mass. Importantly, an inability to increase AT mass by adipocyte hyperplasia may lead to adipocyte hypertrophy and could induce dysfunction of adipose tissue characterized by decreased insulin sensitivity, hypoxia, increased parameters of intracellular stress, increased autophagy and apoptosis and tissue inflammation. As a result, adipocytes and other AT cells release signals (e.g. adipokines, cells, metabolites) resulting in a proinflammatory, diabetogenic and atherogenic serum profile. These adverse signals may contribute to further AT inflammation and secondary organ damage in target tissues such as liver, brain, endothelium, vasculature, endocrine organs and skeletal muscle. Recently, a specific adipocyte volume threshold has been shown to predict the risk for obesity-associated type 2 diabetes.

Most likely, impaired adipocyte function is caused by genetic, behavioural and environmental factors which are not entirely understood. Elucidating the mechanisms of adipocyte dysfunction may lead to the identification of novel treatment targets for obesity and the MetS.

Keywords Adipose tissue dysfunction · Obesity · Metabolic syndrome · Adipocyte · Adipokines · Inflammation

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1 Introduction

Obesity is an important risk factor for the Metabolic Syndrome (MetS), type 2 diabetes, dyslipidemia, fatty liver disease, hypertension, cardiovascular disease and certain types of cancer [1–3]. Activation of the immune system and chronic low-grade inflammation may link excessive fat accumulation to obesity-related MetS and macrovascular complications [1, 2]. The activation of the immune system in obesity is not only reflected by higher circulating concentrations of proinflammatory cytokines, but also by infiltration of macrophages and other immune cells in adipose tissue (AT), liver, muscle and pancreas. Immune cell populations shift towards a proinflammatory profile with a production of pro-inflammatory cytokines, which both affect insulin signaling in peripheral tissues and induce β -cell dysfunction and subsequent insulin secretion defect [2]. It has been recently suggested that the NOD-like receptor family pyrin domain containing-3 (NLRP3) inflammasome senses obesity-associated non-microbial danger signals and contributes to obesity-induced inflammation [4]. NLRP3 inflammasome senses lipotoxicity-associated intracellular ceramide increases to initiate caspase-1 cleavage in macrophages and adipose tissue [4]. Moreover, amyloid deposits in the pancreas have been shown to trigger the NLRP3 inflammasome and may generate IL-1 β [5]. The important role of interleukin (IL)-1 β in the pathogenesis of type 2 diabetes is further supported by clinical trial data demonstrating that an IL-1 blockade by the interleukin-1-receptor antagonist anakinra improves glycemia, insulin secretion and reduced markers of systemic inflammation [6].

At the level of adipose tissue, systemic inflammation may be initiated by dysfunction of adipocytes [2]. Adipocyte dysfunction maybe caused by intracellular “toxins”, such as ceramides or other lipids [4], which accumulate in AT during weight gain. However, not all obese individuals develop chronic systemic inflammation, metabolic or cardiovascular

diseases [7–9]. Therefore, there must be factors, which protect a subgroup of obese individuals against these obesity-related traits. It has been suggested that heterogeneity in body composition, fat distribution and adipose tissue function may be more important to determine the individual cardio-metabolic risk than body fat mass [3, 8–10]. In patients with lipodystrophies, the extent of fat loss correlates with the severity of metabolic complications including diabetes, hypertriglyceridemia, and liver fat content [11]. Moreover, transplantation of AT in rodent models led to beneficial effects on glucose and lipid metabolism [12, 13]. In human studies, the concept that fat distribution may be closer linked to metabolic alterations than total body fat mass is supported by data demonstrating that reducing subcutaneous AT mass by liposuction does not affect metabolic and pro-inflammatory parameters [14]. In contrast, significant beneficial effects on glucose metabolism and insulin sensitivity have been observed following visceral fat reduction in addition to bariatric surgery in obese individuals [15, 16]. These findings suggest that pathomechanisms linking increased AT mass to chronic inflammation and cardiometabolic diseases involve altered ectopic (visceral, liver) fat deposition and impaired adipocyte function [3, 17, 18].

1.1 Adipocyte dysfunction

Weight gain leads to the accumulation of AT by an increase of adipocyte volume (hypertrophy) or number (hyperplasia) [19]. The expansion of AT significantly influences adipocyte biology and subsequently impairs whole-body glucose homeostasis. Hypertrophy of adipocytes is considered a key event associated with a loss of insulin sensitivity both in lean and obese conditions [8, 20–22]. Individuals with larger adipocytes typically have elevated pro-inflammatory factors including leptin, IL-6, IL-8, monocyte-chemotactic-protein-1 (MCP-1) [23], reduced levels of the insulin-sensitivity-related adipokine adiponectin and IL-10 [7, 8, 23], and increased basal and catecholamine-stimulated lipolysis [24] (Table 1). Moreover, studies in mice with an adipose tissue selective deletion of the insulin receptor, revealed an intrinsic heterogeneity of adipocyte size accompanied with distinct changes of both mRNA and protein expression of key molecules in adipocyte biology [25, 26] (Table 1). In obese individuals, adipocyte hypertrophy seems to be associated with deleterious effects on inflammation and metabolism. Quantitative associations have been found between adipocyte volume and insulin resistance and recently an adipocyte volume threshold above which the type 2 diabetes risk increases significantly has been described (Fig. 1) [20]. Interestingly, the concept of an adipocyte size threshold for increased risk of insulin resistance and type 2 diabetes holds true in different populations [20]. However, the size threshold has been shown to be cohort-specific most likely due to both biological

Table 1 Characteristic differences between small and large adipocytes. G-CSF, granulocyte colony-stimulating factor, IL, interleukin; TNF α , tumour necrosis factor alpha; MCP-1, monocyte-chemotactic-protein-1 (modified from [8, 23–26, 32, 72])

Parameter	Small adipocytes	Large adipocytes
Mean diameter	50 μ m	>100 μ m
Insulin stimulated glucose uptake	>5fold	1.5-2fold
Basal triglyceride synthesis	Normal	↑↑
Basal lipolysis	↔	↑
Catecholamine stimulated lipolysis	↔	↑
Leptin secretion (<i>ex vivo</i>)	↔	↑
IL-6 secretion (<i>ex vivo</i>)	↔	↑
TNF α secretion (<i>ex vivo</i>)	↔	↑
IL-8 secretion (<i>ex vivo</i>)	↔	↑
MCP-1 secretion (<i>ex vivo</i>)	↔	↑
IL-10 secretion (<i>ex vivo</i>)	↔	↓
G-CSF secretion (<i>ex vivo</i>)	↔	↑
COL6A3 mRNA	Normal	2.6 fold ↑
Fas, FasL protein	↔	↑↑

variations and technical differences in adipocyte size measurements [20]. Most importantly, having a large adipocyte size was associated with a lower incidence of remission of the insulin resistance state 6 months after bypass surgery even after taking into account established predictors such as baseline glycemic state or diabetes duration [20].

The sequence, how adipocyte dysfunction may contribute to inflammation and metabolic abnormalities could start with

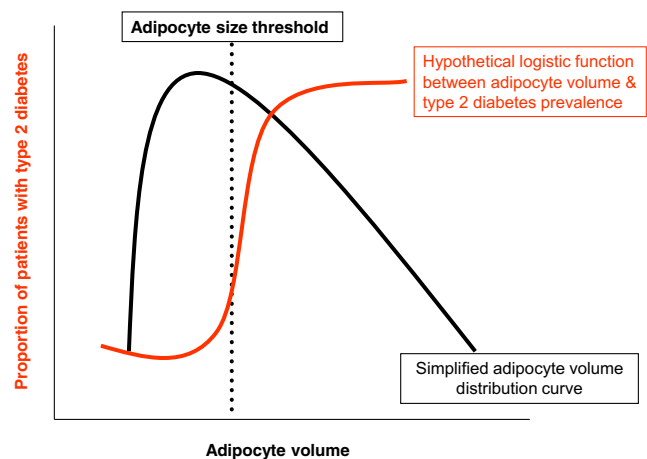


Fig 1 Adipocyte size is linked to the risk of type 2 diabetes. A specific adipocyte volume threshold may predict an increased risk for obesity-associated type 2 diabetes. Such a threshold needs to be established for specific investigation sites and study populations. A simplified adipocyte volume distribution curve (black line) and a hypothetical logistic function between adipocyte volume and prevalence of type 2 diabetes are shown. Noteworthy, a higher adipocyte size prior to significant body weight reduction (e.g. achieved by bariatric surgery) is associated with a lower improvement of insulin resistance. (modified from [20])

adipocyte hypertrophy, which leads to insulin resistant adipocytes with a high lipolytic capacity and secretion of pro-inflammatory and diabetogenic adipokines (Fig. 2). In hypertrophic adipocytes, toxins may accumulate and induce intracellular stress [27, 28], which leads to autophagy as a protective cellular mechanism or apoptosis (Fig. 2) [29–32]. At the tissue level, these mechanisms may contribute to immune cell infiltration of AT, hypoxia, impaired adipogenesis, adverse fat distribution and chronically altered signals from adipose tissue (metabolites, adipokines, cytokines, immune cells). Adverse signals from AT could finally cause secondary organ damage in target tissues such as the brain, liver, skeletal muscle, pancreatic islets and other organs (Fig. 3). In the following review, these mechanisms and the relationship between adipocyte dysfunction, inflammation and the MetS are discussed.

1.2 Adipocyte hypertrophy

Adipocyte hypertrophy could be described by the predominant presence of large, lipid-laden adipocytes with a volume >800pL in AT and correlates with impaired insulin sensitivity, deteriorated glucose and lipid metabolism [3, 33, 34]. Adipocyte hypertrophy maybe the result of an intrinsically low capacity of *de novo* adipogenesis [33]. In contrast, a higher adipocyte generation rate has been shown to lead to hyperplasia of adipocytes [33–35]. The total number of adipocytes is

determined as early as in childhood and adolescence independently of body weight, suggesting that hypertrophy of adipocytes represents the most important mechanism for AT expansion upon weight gain [35]. This conclusion derived from studies involving patients with early-onset (childhood) obesity. Adult individuals who gradually and more slowly increase their body weight over years may initially increase their adipocyte size to a certain threshold before recruiting new adipocytes from precursor cells or mesenchymal stem cells [35]. Adipocyte hyperplasia may at least in part explain the metabolically healthy obese subphenotype [8], because AT hyperplasia has been shown to be protective against lipid as well as glucose/insulin abnormalities in obesity [34]. We and others have demonstrated that the (mean and maximal) adipocyte volume is associated with significantly impaired whole body insulin sensitivity, increased circulating parameters of inflammation and oxidative stress as well as increased number of macrophages within adipose tissue [8, 20, 34, 36–38]. In clinical practice, the differentiation between hypertrophic and hyperplastic obesity is difficult. However, differences in the AT secretome may guide us in the future to diagnose these distinct obesity subphenotypes for a better prediction of the cardiometabolic risk. AT hypertrophy maybe reflected by elevated CrP, IL-6, IL-8, MCP-1, granulocyte colony-stimulating factor (G-CSF), progranulin and chemerin serum concentrations (Table 1) [3, 8].

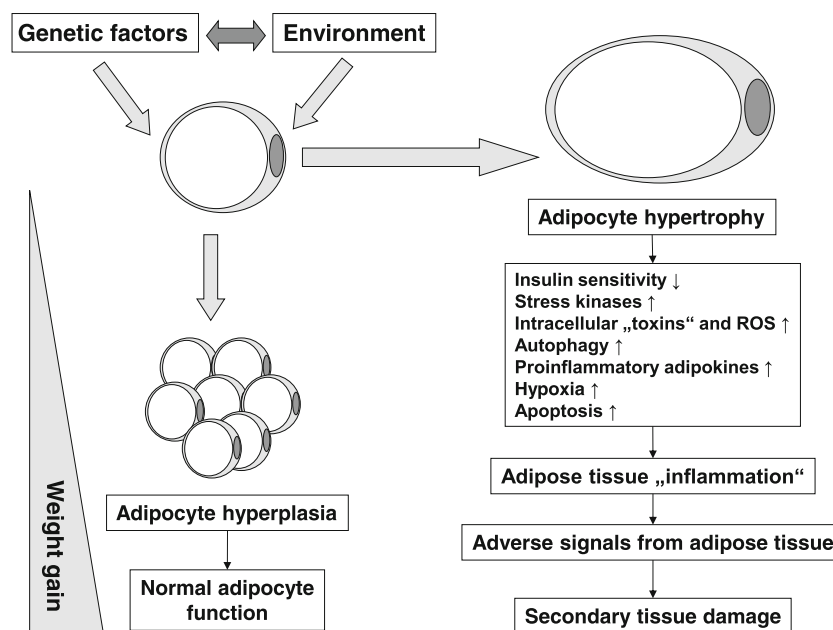


Fig 2 Consequences of adipocyte hypertrophy. With a continued positive energy balance body weight and fat accumulation increase. Most likely due to genetic and environmental factors and their interaction, some individuals may increase the size of adipose tissue depots by both increasing adipocyte size and number (hyperplasia of adipose tissue), which is typically associated with normal adipose tissue function. However, the

majority of patients may respond to the positive energy balance by adipocyte hypertrophy, which is frequently associated with pathogenic factors causing impaired adipose tissue function. As a result, adipose tissue inflammation may develop and could contribute to secondary organ damage via adverse signals from adipose tissue

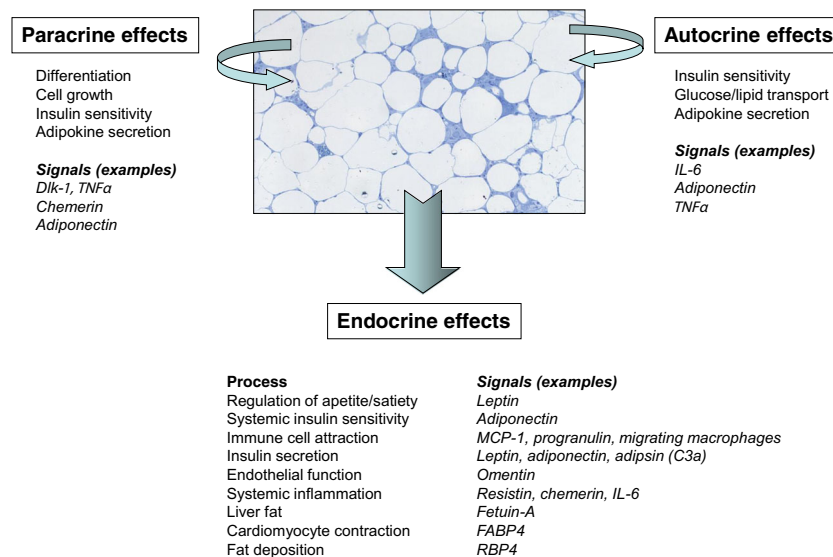


Fig 3 Signals from adipose tissue modulate important biologic processes via autocrine, paracrine and endocrine mode of actions. Adipokines regulate adipogenesis, adipocyte metabolism, immune cell migration into adipose tissue via autocrine and paracrine signalling. In addition, adipokines have endocrine/systemic effects on appetite and satiety 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. IL, interleukin; TNF α , tumour necrosis factor alpha; MCP-1, monocyte-chemotactic-protein-1; FABP4, fatty acid binding protein 4; RBP4, retinol-binding-protein-4 (modified from [3])

1.3 Hypertrophy puts adipocytes under “stress”

The activation of the AT inflammasome plays an important role in the development of insulin resistance [4, 5]. Adipocyte hypertrophy may cause locally and systemically a state of low-grade inflammation which may together with the intracellular accumulation of “toxic” lipids (e.g. palmitate) and advanced glycation endproducts (AGEs) induced IL-1 β secretion via caspase-1 and inflammasome activation [39]. Recently, Esser and colleagues [40] provided important evidence that metabolically healthy obesity is associated with a lower activation of the NLPR3 inflammasome in macrophages of visceral AT and a more favourable inflammatory profile as compared to patients metabolically unhealthy obese individuals.

These recent data are in accordance with the concept that adipocyte hypertrophy, but also ectopic fat deposition and impaired subcutaneous expandability may inflict a variety of stresses on AT [27]. Increased levels of oxidized proteins have been demonstrated in adipose tissue of obese mice, indicating that oxidative stress occurs in adipose tissue [41].

Adipocytes, macrophages, endothelial and other AT cells can activate stress-sensing pathways in response to different stress stimuli, which may lead to cellular malfunction and contribute to inflammation and MetS associated with obesity [27, 28]. Indeed, intracellular kinases that transmit stress response such as p38MAPK and Jun N-terminal kinase (JNK) are upregulated and activated in AT of individuals with visceral fat distribution and adipocyte hypertrophy [28]. The degree of p38MAPK and JNK phosphorylation specifically in

visceral AT correlates with fasting triglycerides, insulin, and glucose [28]. The concept that adipocyte stress may contribute to inflammation and MetS upon weight gain is further supported by studies demonstrating that mice with an AT selective loss of JNK1 are protected against high-fat diet-induced insulin resistance in the liver and increase of inflammatory parameters [42]. The AT JNK1 deficient model further showed that activation of AT stress signaling pathways contributes to the proinflammatory, diabetogenic adipokine secretion pattern [42]. In human studies, we have shown that an activation of the Ask1-MKK4-p38MAPK/JNK stress-sensing pathway correlates with fat accumulation, hypertrophic obesity, AT inflammation and whole-body insulin sensitivity [43]. Different AT stresses may initiate a sequence of events leading to increased immune cell infiltration into adipose tissue [3, 36, 44]. In addition to (hypertrophied) adipocytes, AT macrophages are exposed to the same stressors and may contribute to the link between AT stress response and the risk of obesity induced inflammation and MetS. Supporting this hypothesis, it has been shown, that mice with selective JNK deficiency in macrophages have reduced AT macrophage infiltration and remain insulin sensitive despite high fat diet induced obesity [45].

Importantly, AT macrophages may signal the AT stress and inflammatory status to other organs and could thereby contribute to the inter-organ cross-talk linking obesity to secondary organ failure (Fig. 3). Using live imaging, we recently found AT macrophages emigrate out of so called crown like structures surrounding apoptotic adipocytes to become resident in the interstitium [46]. Moreover, there was a time-

dependent increase in local AT macrophage proliferation and M2 polarization when mice were fed a high-fat diet different [46]. Taken together these data support a model of different recruitment mechanisms for classically activated (M1) and alternatively activated (M2) macrophages in obesity [46].

Obesity, nutrient overload and metabolic abnormalities can initiate or enhance endoplasmic reticulum (ER) stress and activation of the unfolded protein response (UPR) in adipose tissue [27, 47]. ER stress reflects functional overload of this organelle in response to increased protein synthesis, secretion requirements or reduced elimination of misfolded proteins [3, 27]. UPR activation induces transcription of key genes involved in the assembly, folding, modification, and degradation of proteins to alleviate ER stress [3, 48], increase stress kinases activity and proinflammatory cytokine expression [48]. Importantly, elevation in free fatty acids can induce ER stress also in adipocytes thereby linking both overnutrition and increased lipolysis in hypertrophic obesity to adipocyte dysfunction [3, 48].

In obese mice, administration of chemical chaperons, which alleviate ER stress, improved chronic AT inflammation as well as insulin signalling and led to protection against further body weight gain [49]. Taken together, different stresses in AT -including ER stress- play an important pathophysiological role in weight gain and increased nutrient flux induced adipocyte dysfunction.

1.4 Autophagy in adipocytes

Autophagy represents an evolutionary conserved process, which is implicated in the degradation and recycling of damaged organelles, aggregated proteins, and carbohydrates (glycogen) [30, 50]. Autophagy could be considered a cell-protection mechanism against various stresses, induced by damaged organelles for cell survival. Autophagy has been extensively studied in the context of aging, cellular development and cellular defense against pathogens, and is implicated in the pathogenesis of a growing number of human diseases [51]. Moreover, autophagy has been shown to be important for the regulation of lipid metabolism, and pathophysiology of obesity and its related metabolic comorbidities are closely associated with enhanced autophagy [29, 30]. Studies in mice with a deletion of the autophagy gene, autophagy-related 7 (Atg7) specifically in adipose tissue, revealed that autophagy contributes to the regulation of fat mass and the balance between white and brown adipocytes [52]. Atg7 knockout mice are lean, insulin-sensitive, have enhanced metabolic rate, and are resistant to develop obesity [3, 30, 52]. Recently, it has been found that markers of autophagy, such as Atg5, Atg12-Atg5 complex, and the lipidated/cleaved form of LC3 (LC3 II) are up-regulated in human adipose tissue in obesity, especially in visceral fat [29]. Moreover, we have shown that insulin resistance in adipose tissue of obese Wistar Ottawa Karlsburg

W rats is associated with up-regulation of different autophagy markers in visceral and subcutaneous fat depots [53]. Increased expression of autophagy genes correlates with the degree of obesity, visceral fat distribution, and adipocyte hypertrophy both in humans and rodent models [29, 53]. The activation of autophagy may occur together with the development of insulin resistance, but could precede the occurrence of obesity-associated morbidity [3]. Therefore, it is still debated whether autophagy may either protect against obesity associated adipocyte dysfunction or could be a symptom of impaired AT function.

1.5 Apoptosis of adipocytes

Apoptosis of adipocytes is increased in AT from both mice with diet-induced obesity and obese humans [31, 32, 54]. Adipocyte death defines macrophage localization and function in AT and may represent an early event in immune cell attraction and AT inflammation [46, 55]. In mice, genetic inactivation of Bid, a key pro-apoptotic molecule, significantly reduced adipocyte apoptosis prevented immune cell infiltration of AT and protected against the development of whole body insulin resistance [54]. Moreover, adipocyte specific deletion of the death receptor Fas (CD95), a member of the TNF receptor family, which plays a key role in promoting apoptosis, improves insulin sensitivity and reduces hepatic steatosis as well as AT inflammation in high fat diet-fed mice [56]. On the contrary, Fas activation in adipocytes did not only stimulate the production of proinflammatory cytokines, but also interfered with insulin-stimulated glucose uptake and induced basal lipolysis further suggesting a role of Fas in adipocyte function [57, 58].

To extend these findings in rodent models to humans, we recently investigated expression of Fas and FasL in human AT [32]. We found significant relationships between AT *Fas* expression and adipocyte apoptosis, parameters of obesity, fat distribution, AT function, and insulin sensitivity [32]. In isolated adipocytes *Fas* expression was significantly higher in obese compared to lean individuals and in patients with type 2 diabetes compared to normal glucose tolerant healthy controls [32]. Very recently, it has been shown that two other apoptosis-related proteins, the death ligand TNF-related apoptosis-inducing ligand (TRAIL) [59] and B-cell lymphoma 2 (*bcl-2*) [60], play a previously unrecognized role in regulating adipocyte metabolism. Differential regulation of these molecules may contribute to the observation that mature adipocytes are less sensitive to apoptotic stimuli as compared to preadipocytes [60]. Moreover, a shift in the balance of proapoptotic and anti-apoptotic molecules during adipogenesis, which results in higher apoptosis resistance may represent an intrinsic mechanism for a preserved AT function. Taken together, these data suggest that adipocyte apoptosis is a key initial event that contributes to macrophage infiltration into

adipose tissue and insulin resistance associated with obesity in both mice and humans. Targeting mechanisms which increase AT apoptosis could be a novel approach for the prevention and treatment of obesity and its related metabolic diseases.

1.6 From adipocyte dysfunction to impaired function of adipose tissue

Genetic and environmental factors as well as their interaction may play a role both as causative factors for obesity development itself and for the dissociation into subphenotypes of hyperplastic and hypertrophic fat accumulation (Fig. 2). However, so far genotype-environment interaction studies have provided only limited advancement in our knowledge how genetic and environmental factors interact to determine distinct human phenotypes. Recently, the hypothesis that genotype—environment interaction may contribute to the variance of the supra-iliac skinfold thickness—which is related to abdominal obesity - has been tested in 7,170 unrelated Korean individuals on 326,262 autosomal single nucleotide polymorphisms [61]. Importantly, the authors demonstrate that certain genotypes could have different effects on this surrogate parameter of abdominal obesity in different environmental settings (rural compared to urban areas) [61]. Supporting our hypothesis that gene-environment interactions may also contribute to the distinction between hyperplastic and hypertrophic obesity subphenotypes (Fig. 2), the recent study from Korea revealed that gene clusters responsible for cell-cell and cell-extracellular matrix interactions were enriched significantly for the genotype-environment interaction [61].

Environmental factors may either directly, in genetically susceptible individuals or via epigenetic alterations, contribute to individual differences in fat distribution as well as obesity subphenotypes [62]. Among such environmental factors, food contaminants, particularly those which are considered endocrine disruptors, may cause ectopic fat accumulation and adipocyte hypertrophy [62]. There is also good evidence in humans that sugar-sweetened beverages promote the accumulation of visceral fat [63]. Moreover, mental stress, psychosocial and socioeconomic factors, depressive and anxiety traits, alcohol and smoking may have significant influences on body fat distribution and at least hypothetically adipocyte dysfunction [62, 64].

Hypertrophic obesity (Fig. 2) may initiate a sequence of pathogenic factors leading to AT dysfunction and subsequent effects on other tissues.

The main function of AT is the storage of fat under conditions of excess calories and their release in periods of fasting. In addition, AT plays a role in thermoregulation, mechanical organ protection and as an endocrine organ [3]. In 2014, the research community celebrates the 20th anniversary of the discovery of leptin [65]. Preceding this important discovery, adipose tissue has been identified as an endocrine organ and

adipsin/complement factor D was the first adipokine described [66, 67], but it took ~20 years to understand how adipsin links adipocyte function to β cell physiology [68]. Adipsin generates the peptide C3a, which is a potent insulin secretagogue and may explain the beneficial effects of adipsin on insulin secretion and whole body glucose metabolism [68]. Since the discoveries of leptin and adipsin, we have learnt that adipokines regulate important biological processes in target organs including the brain, liver, skeletal muscle, vasculature, heart, immune system and pancreatic islets (Fig. 3). Altered adipokine secretion may contribute to impaired regulation of appetite and satiety, fat distribution, insulin secretion and sensitivity, energy expenditure, endothelial function, inflammation, blood pressure, and hemostasis [3]. Therefore alteration in adipokine secretion may link obesity to its inflammatory, metabolic and cardiovascular comorbidities [3]. Because adipokines also act in an autocrine and paracrine manner they could contribute to the development of AT dysfunction initiated from disturbances of individual adipocytes. In addition to changes in adipokine secretion, impaired expandability of subcutaneous AT, ectopic fat deposition and hypoxia may contribute to adipose tissue inflammation or dysfunction.

1.7 Impaired subcutaneous AT expandability

In principle, subcutaneous (SC) AT has a higher capacity to expand its capillary network than AT in visceral depots or organ structure AT, but with the gain of fat mass this capacity decreases [69–72]. This decrease correlates with insulin resistance [69]. It has been therefore suggested that impaired SC vascularisation capacity may contribute to lower SC AT expandability and metabolic diseases [69]. In addition, adipogenesis may play a pivotal role in SC expandability [69, 70]. Differences of adipogenic capacity of progenitor cells from different depots and the loss of adipogenic potential that occurs during the course of obesity might contribute to adipose tissue dysfunction [70]. Studies in different animal models suggest that expansion of adipose tissue requires angiogenesis [3, 71]. It has been demonstrated that SC AT has a higher capillary density per adipocyte and higher angiogenic growth capacity compared with visceral tissue [69]. The results support the existence of intrinsic differences in angiogenic capacity between different fat depots [69].

The inability of SC AT to expand proportional to caloric excess represent an important junction in the development of AT dysfunction and fat accumulation in ectopic depots such as omentum, mesenterium, liver and other organs [3, 70–72]. Even within AT impaired SC AT expandability may lead to changes in the extracellular tissue matrix which could be considered as adipose tissue fibrosis [73, 74]. In line with that, increased expression of the important matrix protein collagen 6A (COL6A) has been associated with adipocyte hypertrophy, insulin resistant obesity and metabolic abnormalities [75].

Adipose tissue has a remarkable capacity to expand. However, under conditions of severely impaired AT expandability such as genetic syndromes of lipodystrophies lipids accumulate in the liver and muscle and insulin resistance, dyslipidemia and endothelial dysfunction develop prematurely [3, 11]. In analogy to the lipodystrophy model, deterioration of glucose and lipid metabolism maybe the result of an impaired capacity to store fat in the “benign” SC fat depot, which could be considered as an innocent metabolic sink [76]. As SC AT reaches its storage capacity, excess calories are being redirected to other depots and lead to ectopic fat accumulation. Interestingly, metabolically healthy obese individuals seem to have a preserved SC AT expandability and are (therefore?) protected against early manifestation of obesity-associated metabolic diseases [8].

The SC AT expandability concept has been recently challenged by systematic 8 weeks overfeeding (>40 % of baseline requirements) experiment in 29 overweight volunteers [77].

Contrary to expectations, this study demonstrated that substantial weight gain together with the occurrence of larger adipocytes did not promote ectopic lipid accumulation, whereas smaller fat cells were associated with a worsened metabolic response to overfeeding [77].

1.8 Adverse fat deposition

Central body fat distribution has been shown to better predict obesity-related cardiometabolic diseases than whole body fat mass or body weight [78–80]. Visceral abdominal obesity has been shown to be more strongly associated with adverse outcomes of obesity than peripheral or subcutaneous obesity [78–81]. These different associations maybe due to intrinsic properties of visceral and SC fat depots with regard to insulin sensitivity, lipolytic activity, angiogenic potential, cellular composition and expression of key genes of adipocyte biology [3]. In addition, the anatomical site of visceral fat could contribute to an increased cardiometabolic risk, because this depot drains into the portal vein making the liver a target of undiluted metabolites, cytokines and adipokines released from visceral fat [62]. Fat distribution is influenced by genetic factors. Waist to hip ratio (WHR), a surrogate parameter for central fat distribution, has a significant heritability of up to 60 % [reviewed in 62]. Twin and population studies have revealed that both body mass index (BMI) and WHR are heritable traits, with genetics accounting for 25–70 % of the observed variability [62, 81, 82]. Although there have been considerable recent insights into the control of appetite and energy expenditure as factors contributing to obesity, little is known about the genetic factors determining of adipocyte number or differences in body fat distribution. By using the latest advances in high-throughput technologies as well as statistical approaches for well-powered genome-wide association studies (GWAS), numerous novel genes or loci have

been recently identified (e.g. MC4R, TFAP2B, RSPO3, VEGFA, TBX15–WARS2, NFE2L3, GRB14–COBLL1, DNM3–PIGC, ITPR2–SSPN, LY86, HOXC13, ADAMTS9, ZNRF3–KREMEN1, NISCH–STAB1, CPEB4 [62, 83, 84]. A recent GWAS including up to 263,407 individuals of European ancestry identified associations between WHR and genetic variants in the genes RSPO3, LY86, LYPLAL1 and COBLL1 [83]. Altogether, the GWAS findings indicate a strong genetic background for the regulation of body fat distribution, independently of overall obesity. Interestingly, major patterns of fat distribution may have a developmental genetic origin [85]. Both in humans and mouse models, specific developmental genes show a fat-depot specific expression pattern which correlates with either predominantly visceral or SC fat accumulation and metabolic traits [85]. However, the function of these genes and the mechanisms underlying their association with adverse fat distribution are still not entirely understood. Taken together, the functional characterization of previously unrecognized fat distribution genes identified by recent GWAS will be crucial for unraveling the complex etiology of obesity-related complications and might even provide novel strategies for the treatment of obesity and the MetS.

1.9 “Inflammation” of adipose tissue

The term inflammation of AT is now frequently used as a synonym for AT dysfunction, although this is not necessarily describing the alterations in AT correctly. Mechanisms including adipocyte hypertrophy, AT stresses and apoptosis which cause pro-inflammatory adipokine secretion may lead to the attraction of pro-inflammatory immune cells into adipose tissue causing chronic, low-grade inflammation [36, 44, 55]. Increased number of AT macrophages mainly in visceral depots is associated with adipocyte hypertrophy, insulin resistance, activation of stress-signalling pathways, increases in autophagy and apoptosis [reviewed in 3]. Mechanisms of increased macrophage numbers in AT include both recruitment of circulating monocytes by chemoattractant proteins including MCP-1, chemerin, progranulin, colony stimulating factor –1 (Csf1) and local proliferation [3, 46]. Importantly, AT macrophages switch their phenotype towards a pro-inflammatory subtype, a process in which cells of the adaptive immune system are involved [86]. In response to excessive nutrient intake, T-cell phenotype changes and recruitment of B cells and T cells may precede AT macrophage infiltration [86]. The higher number of immune cells in visceral AT suggests that antigens derived from the gut may contribute to T-cell activation and recruitment into visceral adipose tissue [86]. Within adipose tissue, immune cells are important sources for cytokine and chemokine production, which maintain or worsen both locally and systemically a low-grade inflammation [3]. Noteworthy, obesity and inflammation are highly

integrated processes in the pathogenesis of insulin resistance, diabetes, atherosclerosis, and non-alcoholic fatty liver disease [87]. Macrophage infiltration into adipose tissue increases proportionally with increased BMI, body fat mass and adipocyte hypertrophy and represents a reversible process in obese patients losing weight [3, 36, 44].

Hypoxia represents an additional and independent mechanism for both the development of AT dysfunction and the recruitment of macrophages into AT [3, 88]. It has been shown that hypoxia inducible factor-1 α (HIF-1 α): a transcription factor strongly induced by hypoxia is over-expressed in adipose tissue of obese patients [36]. Moreover, recently, increased BMI in adults has been associated with increased methylation at the HIF3A locus in blood cells and AT, suggesting that perturbation of hypoxia inducible transcription factor pathways could have an important role in the response to increased body weight [89].

In summary, AT inflammation maybe an important pathomechanism contributing to systemic inflammation and metabolic disturbances associated with increased food intake and body weight gain.

1.10 Adipokines may link adipocyte dysfunction to the metabolic syndrome

Via the secretion of a pro-inflammatory adipokines and emigrating immune cells, “inflamed” AT may signal its functional status to other organs (Fig. 3). After the discovery that adipose tissue is an active endocrine organ, many AT secreted molecules have been identified. Today, at least 600 bioactive factors are considered as adipokines [90, 91]. The function, signalling and mode of action of many of these more recently discovered adipokines are still elusive.

Adipokines may play specific roles in immune response (e.g. adiponectin/complement factor D, acylation-stimulating protein, serum amyloid A3, interleukins) and inflammation (e.g. IL-1 β , IL-6, IL-8, IL-10, CrP, MCP-1, osteopontin, progranulin, chemerin), glucose metabolism (e.g. leptin, adiponectin, dipeptidyl peptidase-4, fibroblast growth factor 21, resistin, vaspin), insulin sensitivity (e.g. leptin, adiponectin, chemerin), hypertension (e.g. angiotensinogen), cell adhesion (e.g. PAI-1), vascular growth and function (e.g. VEGF), atherosclerosis development (e.g. cathepsins, apelin), adipogenesis and bone morphogenesis (e.g. BMP-7), growth (e.g. IGF-1, TGF β , fibronectin), lipid metabolism (e.g. CD36), regulation of appetite and satiety (e.g. leptin, vaspin) and other biological processes [92, 93]. In parallel with AT expansion and obesity development, adipokine secretion is significantly altered towards a diabetogenic, proinflammatory, and atherogenic adipokine pattern [92, 93]. An adipokine pattern with low circulating adiponectin and high serum concentrations of chemerin, IL-6, TNF α , progranulin, RBP4,

fetuin-A, CrP, DPP-4 and others may reflect AT dysfunction [3, 92–94].

Targeting or using adipokine-based mechanisms to treat obesity and diseases which are caused by a positive energy balance is therefore a promising strategy. Adipokine research of the past 20 years has led to clinical application of the leptin analogue metreleptin as a pharmacotherapy in individuals with congenital leptin deficiency and lipodystrophies [92]. Other adipokine-based treatment strategies using for example adiponectin, FGF21 or BMP7 may follow leptin in the path of drug discovery. In summary, adipokines may open exciting new treatment opportunities for diseases with several unmet clinical needs.

2 Summary

Adipocyte hypertrophy and dysfunction belongs to the primary defects in obesity and may link excessive fat storage to chronic systemic inflammation, metabolic and cardiovascular diseases. With increasing body weight upon excessive energy intake, AT dysfunction may develop and seems to determine the individual risk to develop metabolic and cardiovascular comorbidities. The inability to store excess calories in SC AT depots may represent a critical node in the development of subsequent ectopic fat deposition in visceral depots, the liver and other cell types. Adipocyte dysfunction may therefore initiate a sequence of pathomechanisms including cellular insulin resistance and increased lipolytic capacity, intracellular accumulation of toxic molecules, activation of stress pathways, visceral (ectopic) fat accumulation, changes in the cellular and intracellular matrix composition of AT, increased number of immune cells within AT, increased autophagy and apoptosis, AT extracellular matrix changes (AT fibrosis), alterations in AT mRNA and protein expression patterns.

As a consequence, adipocyte dysfunction contributes to a proinflammatory, atherogenic, and diabetogenic state and maybe mechanistically linked to the development of chronic inflammation, insulin resistance, the MetS and other obesity associated disorders. Targeting adipocyte dysfunction maybe a promising approach to prevent or treat obesity related diseases in the future.

Conflict of interest No potential conflict of interest relevant to this article needs to be reported.

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