Impact of visceral adipose tissue on liver metabolism
Part I: Heterogeneity of adipose tissue and functional properties of visceral adipose tissue

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Abstract
Excess visceral adipose tissue is associated with anomalies of blood glucose homoeostasis, elevation of plasma triglycerides and low high-density lipoprotein cholesterol that contribute to the later appearance of type 2 diabetes and cardiovascular syndromes. Visceral adipose tissue releases a large amount of free fatty acids and hormones/cytokines in the portal vein that are delivered to the liver, and interact with hepatocytes and various immune cells in the liver. The functional characteristics of visceral adipose tissue will be compared with subcutaneous adipose tissue to clarify the major mechanisms affecting free fatty acid metabolism and cytokine production.

Résumé

Un excès de tissu adipeux viscéral est associé à des perturbations de l’homéostasie glucidique, une augmentation des taux de triglycérides plasmatiques et une baisse de la fraction du cholestérol contenu dans les lipoprotéines de haute densité qui vont accroître le risque d’apparition ultérieure d’un diabète de type 2 et de syndromes cardiovasculaires. Le tissu adipeux viscéral libère des acides gras libres et des hormones/cytokines dans la veine porte qui seront directement délivrés au foie. Ces diverses productions vont affecter les fonctions des hépatocytes et des cellules immunitaires présentes dans le foie. Les caractéristiques fonctionnelles du tissu adipeux viscéral seront comparées à celles du sous-cutané, afin de préciser la spécificité des mécanismes qui interviennent dans le contrôle de la production des acides gras libres et des cytokines par le tissu adipeux viscéral.

Keywords: Adipocyte; Obesity; Visceral adipose tissue; Fatty acids; Adipokines; Review
Mots clés : Adipocyte ; Obésité ; Tissu adipeux viscéral ; Acides gras ; Adipokines ; Revue

Abbreviations: ASP, acylation-stimulating protein; AT, adipose tissue; BMI, body mass index; CRP, C-reactive protein; ChREBP, carbohydrate response element-binding protein; ER-α and ER-β, oestrogen receptor-α and -β; FABP, fatty acid-binding protein; FGF, fibroblast growth factor; HSL, hormone-sensitive lipase; 11β-HSD1, 11β-hydroxysteroid dehydrogenase-1; IFN, interferon; IL-1, -6, -8, -10, interleukin-1, -6, -8, -10; IL-1Ra, interleukin-1 receptor antagonist; LPL, lipoprotein lipase; LPS, lipopolysaccharide; MCP-1, monocyte chemoattractant protein-1; MIP-1α, macrophage-inflammatory protein-1α; FFA, free fatty acids (nonesterified fatty acids); PAI-1, plasminogen activator inhibitor-1; PBEF, pre-B-cell colony-enhancing factor; PEPCK, phosphoenolpyruvate carboxykinase; RBP-4, retinol-binding protein-4; SCAT, subcutaneous adipose tissue; SVF, stromal-vascular fraction; T2D, type-2 diabetes; TG, triglycerides; TGF-β, transforming growth factor-beta; TNF-α, tumor necrosis factor-alpha; sTNFR2, soluble type-2 TNF-α receptor; VAT, visceral adipose tissue; VEGF-A, vascular endothelial growth factor-A.

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Obesity is a risk factor for numerous diseases such as type 2 diabetes (T2D), hypertension and cardiovascular events, including myocardial infarction and stroke. The anatomical distribution of fat deposition has a marked impact on the level of risk. Abdominal, or male, obesity, in particular, sharply raises the risk of insulin resistance, T2D, and further risk factors of metabolic and cardiovascular diseases [1]. Patients with abdominal obesity due to adipose tissue (AT) accumulation in the upper part of the body have more extensive visceral AT (VAT) [1]. The insulin resistance, or metabolic syndrome, that precedes T2D onset is usually associated with dyslipidaemia. Patients with excess VAT are characterized by anomalies of blood-glucose homeostasis, elevated plasma triglycerides (TG) and low levels of high-density lipoprotein (HDL) cholesterol. These disorders are often associated with a prothrombotic and proinflammatory state that further contributes to the later appearance of cardiovascular syndromes [1,2]. It has been shown that VAT releases a large amount of free fatty acids (FFA) and hormones/cytokines in the portal vein that are then directly delivered to the liver. The anatomy of the liver also provides a mechanism by which the portal and arterial circulations can interact with both the parenchyma (hepatocytes) and immune-system cells that are also located within the liver.

We discuss here the functional characteristics of VAT in comparison to the subcutaneous fatty tissue to clarify the major mechanisms regulating FFA release and cytokine production.

1. Heterogeneity of the distribution of fat deposits

Abdominal fat is composed of distinct anatomical components: subcutaneous AT (SCAT) contains superficial and deep layers separated by superficial fascia (Scarpa’s fascia) and intra-abdominal fat, located in intra- and retroperitoneal deposits. VAT, predominantly comprising omental and mesenteric deposits, represents 10–20% of total body fat in thin and obese men, and 5–10% of total body fat in women. For the same body mass index (BMI), or fat mass, VAT accumulation is greater in men than women (after correction for differences in total adiposity). Subcutaneous deposits represent 80% of a healthy individual’s total fat mass. A subject with a normal BMI and extensive VAT has a greater risk of developing the metabolic syndrome than a more obese individual with less VAT accumulation.

2. Questions and hypotheses to explain metabolic anomalies associated with extensive VAT

Why is VAT accumulation associated with a more deleterious metabolic profile than fat accumulations at other, subcutaneous sites? Is the extent of VAT a causal factor or simply a marker of an abnormal metabolic profile? Which contributes more specifically to the development of metabolic anomalies associated with visceral obesity: VAT or visceral adipocytes? Is the role of VAT accurately estimated, or overestimated because of its weak quantitative impact on total fat mass?

Two schools of thought compete in trying to explain the glucose intolerance, insulin resistance and lipid abnormalities associated with visceral obesity. The first and older explanation is the “portal hypothesis”, initially advanced by Björntorp [3]. This favours the metabolic role of VAT, the inherent dangers of which are its drainage by the portal vein and its abundant (or excess) delivery of FFA to the liver, with enhanced lipolytic activity due to lipolysis of VAT adipocytes. Plasma FFA concentrations are always elevated in obese and T2D patients [4]. VAT-derived FFA alters several liver functions (discussed in detail in part II) (Fig. 1).

The second hypothesis confers major roles to a wide variety of lipid and peptide processes (tumour necrosis factor-α [TNF-α], interleukin-6 [IL-6], interleukin-8 [IL-8], leptin, resistin, adiponectin released within VAT). Expression of these peptides, which can be emptied into the portal vein with those derived from VAT, varies with distribution of the fat mass. They also have multiple effects, as discussed in recent reviews [5,6].

Obesity is now considered a proinflammatory state, leading to chronic immune-system activation with subacute progression. Indeed, the obese have elevated plasma levels of various biological markers of inflammation [7,8], including acute-phase proteins such as C-reactive protein (CRP), IL-6, TNF-α, α1-acid glycoprotein, IL-1 receptor antagonist (IL-1Ra), acute-phase serum amyloid A (SAA), retinol-binding protein (RBP)-4, haptoglobin, plasminogen activator inhibitor-1 (PAI-1) and metallothionein. Adipocytes such as monocytes/macrophages secrete some of these markers. Macrophages accumulate in the AT of humans and rodents, and their density is strongly increased in the obese [9–12]. The beneficial effects of weight loss on the complications of obesity are associated with diminished production of proinflammatory factors and enhanced synthesis of anti-inflammatory molecules [12,13].

What is the pathophysiological implication of monocytic infiltration of AT in the obese for the pathogenesis of the metabolic and endocrine disorders observed? What are the factors or cellular processes leading to the retention and subsequent accumulation of macrophages in AT in the obese? What role does VAT play in the accumulation of macrophages that produce inflammatory molecules in situ and which are also delivered to the liver? The results of two recent reports drew attention to monocyte chemoattractant protein-1 (MCP-1), which is produced predominantly by macrophages and endothelial cells and, to a lesser extent, by adipocytes. MCP-1, known to be a potent chemoattractant for monocytes, participates in macrophage infiltration of mouse AT and appears to be responsible for the insulin resistance and steatosis observed in obese rodents. Knockout of the mouse gene encoding the MCP-1 receptor (C-C chemokine receptor-2, CCR2), or its blockade by a pharmacological agent, limited macrophage infiltration of AT and insulin resistance induced by a high-fat diet [14]. However, these observations remain to be confirmed in humans. The relative contributions of adipocyte-secreted adipokines and adipose tissue-infiltrated macrophages in the initiation and maintenance of a chronic subinflammatory state, and the development of insulin resistance and T2D, remain to be clarified [15].

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Fig. 1. Working hypothesis for the mechanism(s) involved in the privileged visceral adipose tissue-liver interactions leading to the appearance of obesity-related diseases. Major factors involved in the control adipocyte metabolism and adipose tissue secretions are depicted. Putative incidence of adipose tissue productions on liver dysfunctions is pinpointed.

Visceral fat deposits only represent 5 to 20% of body fat mass. Visceral adipose tissue contains visceral adipocytes, mesothelial cells (specific of the omentum) and a number of different fat cell types in the stroma-vascular fraction (SVF) (that is macrophages, lymphocytes [T cells], microvascular endothelial cells and adipose-derived stem cells). Most of these cell types possess secreting activities which largely remain to be identified but mainly quantified. Until now, the interleukin IL-6 is the unique factor which has been found to be increased in the portal vein of obese patients. Other adipokines, growth factors, chemokines and interleukins secreted by visceral fat deposits are still poorly characterized in human obesity. It is essential to delineate those which appear in portal vein from those devoted to paracrine/autocrine effects. Non-esterified fatty acids (FFAs) are directly delivered by visceral adipocytes to liver and impact on a number of metabolic parameters (see part II). Note that the upper subcutaneous fat depot is also a noticeable site of FFA release. All the abbreviations are defined in the text.

11β-HSD1, type 1, 11β-hydroxysteroid dehydrogenase (in humans, converts the inactive 11-keto metabolite, cortisone into the active glucocorticoid cortisol). (↑) increased (activity or secretion), (↓) decreased (activity or secretion).

3. VAT morphological and proliferative characteristics

In general, visceral adipocytes are smaller than subcutaneous adipocytes in both men and thin women, although this size disparity is lost in obese men, but persists in obese women. In addition to mature adipocytes, visceral fat deposits, like subcutaneous ones, contain a highly heterogeneous cell population that constitutes the stromal-vascular fraction (SVF) of AT (preadipocytes, microvascular endothelial cells, mastocytes, fibroblasts, monocytes/macrophages, progenitor cells of bone-marrow origin, leukocytes and granulocytes). Macrophages are more abundant in VAT than in SCAT [11,16–18].

The abundance of adipocyte precursors (preadipocytes), and their proliferative and differentiation capacity, play determinative roles in the distribution of VAT. Depot-specific variation has been reported in fat-tissue cell dynamics [19]. For example, the results of studies in vitro show that the replicative capacity of omental preadipocytes is inferior to those of their subcutaneous counterparts. The proliferative ability of subcutaneous adipocyte precursors disappears with age, whereas that of omental fat precursors appears to be less affected by ageing. In contrast, the differentiation capacity of preadipocytes at both locations is apparently the same. It is difficult to extrapolate these in vitro observations to the actual biology of these fat deposits. Indeed, the survival and evolution of SVF cell types depend on the culture techniques used [20,21]. Adipocyte progenitors are included in the SVF fraction, identified by CD34+/CD31 surface markers, and have properties that distinguish them from adult mesenchymatous cells and hematopoietic stem cells [22]. A recent study has shown that the proportion of subcutaneous SVF cells that are committed preadipocytes is reduced with obesity [23]. This may be explained by either a greater recruitment of preadipocytes to adipogenesis or a greater preadipocyte apoptosis, depending upon the obesity phenotype. In rats, ageing results in a paradoxical susceptibility of fat-cell progenitors to lipotoxicity [24].
4. Metabolic characteristics of visceral adipocytes

4.1. TG synthesis, lipoprotein lipase activity and acylation-stimulating protein

Although key enzymes in the synthesis of fatty acids have been identified in human adipocytes, their contribution to lipogenesis remains minimal. More fatty acids are synthesized in VAT adipocytes [25]. Indeed, in humans, adipocytes synthesize the majority of their TG from glucose uptake and FFA derived from lipoprotein lipase (LPL) activity. LPL activity rises in parallel with increases in adipocyte size. In women, VAT has lower LPL activity than abdominal, gluteal and femoral SCAT, although these differences regress at the menopause. LPL-activity disparities are less evident in men. Insulin stimulates LPL activity, glucose transport and TG synthesis; it has a greater impact on LPL activity in SCAT than in VAT [26]. Basal glucose-transport activity and insulin-stimulated glucose transport are more intense in omental than subcutaneous adipocytes (with no modification of their sensitivity to insulin). Insulin-stimulated glucose transport is two times higher in omental than subcutaneous adipocytes from the same patient [27]; this effect is also seen in the insulin-signaling pathways in adipocytes derived from the differentiation of VAT and SCAT preadipocytes in prepubertal children. The level of glucose-transporter GLUT4 mRNA expression is elevated in mature adipocytes in the VAT of adults (quadrupled in VAT compared with SCAT) [28,29]. The same holds true for the expression of insulin-signaling transcription proteins [30]. These in vitro observations have been supported by the use of \( ^{18}\)F-2-fluoro-2-deoxy-D-glucose positron emission tomography (PET), which reveals that insulin-induced glucose uptake is greater in VAT than in SCAT [31]. The enhanced glucose uptake by omental adipocytes facilitates TG neosynthesis to counterbalance the accrued lipolysis observed in VAT. Based on lipolysis data (see below) and the effects of insulin on glucose uptake, the turnover of TG reserves appears to be greater in VAT.

Fatty-acid uptake activity and the level of acyl-coenzyme A synthase activity contribute to adipocyte synthesis of TG. The published fatty-acid compositions of subcutaneous and omental adipocytes have been inconsistent, as they are partly dependent on food intake and on the nutritional habits of the populations studied. Omental adipocytes have an enhanced ability to incorporate fatty acids. More tritiated olein is taken up by VAT than SCAT in both obese and thin subjects. In contrast, fatty-acid-binding protein (FABP) is more strongly expressed in subcutaneous than omental adipocytes in obese individuals [32].

Acylation-stimulating protein (ASP), called C3adesArg, is a circulating protein produced by adipocytes from adipin. Secreted after a meal, ASP stimulates TG synthesis and plays a role in postprandial TG clearance. ASP binds to receptors on the adipocyte plasma membrane and stimulates diacylglycerol-acyltransferase activation [33]. ASP-binding capacity and affinity are weaker in VAT than in SCAT. The physiological role of this protein remains to be elucidated.

Adipogenesis and the expression of numerous enzymes controlling adipocyte metabolism are regulated by the coordinated expression of many transcription factors, of which the best known are sterol regulatory element-binding protein-1c/ADD1 (SREBP-1c/ADD1), peroxisome proliferator-activated receptors (PPAR\(\alpha\), \(-\beta/\gamma\) and \(-\gamma_1/\gamma_2\)) and CCAAT/enhancer-binding protein-\(\alpha\) (C/EBP\(\alpha\)). Little work has been done on regional differences in the expression and activity of these factors in humans. The lowest SREBP-1c mRNA levels are found in omental fat (compared with abdominal and femoral SCAT). No differences are linked to gender. SREBP-1c mRNA levels are lower in the obese and increase after weight loss.

4.2. Lipolysis and mobilization of lipids

The essential metabolic differences between subcutaneous and visceral adipocytes concern modulation of their responses to catecholamine- and insulin-induced lipolysis. These hormones potentely influence FFA turnover in the different fat deposits, and orchestrate the different contributions of the various fat deposits to FFA homoeostasis. The AT-dependent effects of other lipolytic agents, such as growth hormone, natriuretic peptides and IL-6, remain poorly understood.

Lipolysis rates, both in vitro and in vivo, depend on the activity of adipocyte lipases, and the functional equilibrium between the efficacy of lipolytic and antilipolytic regulators. Basal or spontaneous lipolysis is lower in omental adipocytes than in their subcutaneous counterparts in healthy and obese subjects. The increased basal lipolysis seen in subcutaneous adipocytes could reflect their FABP content, which facilitates the export of fatty acids generated by lipolysis.

4.2.1. Effects of insulin

Insulin actions on lipolysis and reesterification of FFA are lower in omental adipocytes. In vitro studies revealed that several insulin-signal transduction molecules are affected [30,34]. In the absence of information on the expression of proteins implicated in the control of lipolytic and antilipolytic pathways (protein kinase-A, phosphodiesterase-3B, perilipin, and lipases such as adipocyte TG lipase [ATGL] and hormone-sensitive lipase [HSL]), it is not possible to interpret these disparities. Note, however, that the differences observed in vitro were confirmed in vivo. VAT is more resistant to the antilipolytic effect of insulin than is femoral or non-VAT AT in humans [35].

4.2.2. Effects of catecholamines

The sympathetic nervous system plays an essential role in the regulation of lipolysis and lipid mobilization in humans [36,37]. Catecholamines act simultaneously on adipocytes and AT vascularization by controlling local blood flow. They also modulate the secretion of the antilipolytic hormone insulin. In normal subjects, catecholamines have a more potent impact on visceral adipocytes than on their subcutaneous counterparts. This difference persists in differentiated preadipocytes in vitro [38]. In contrast, although the results of several studies showed that a \(\beta\)-adrenergic-receptor agonist exerted equivalent effects on adipocytes at various sites in nonobese individuals, a recent
investigation found that the β-adrenergic response was more intense in VAT adipocytes in the obese. In-depth analysis of the signaling pathways revealed no alteration of the postreceptor stages. The discrepant responses were found to be due to the expression of β- (mainly β₁ and β₂) and α₂A-adrenergic receptors in the different deposits. VAT adipocytes have a weaker α₂A-adrenergic response (and low α₂A-adrenergic-receptor density), which explains the potent lipolytic efficacy of catecholamines. In women, subcutaneous gluteal and femoral adipocytes are characterized by their strong α₂A-adrenergic response (and high α₂A-adrenergic-receptor density) associated with a low density of β-receptors[39]. Abdominal subcutaneous adipocytes in obese men have characteristics similar to those of femoral subcutaneous adipocytes in women [40].

With age and depending on the degree of obesity, this functional balance can change and lead to perturbations of the downstream receptor pathways such as diminished expression of HSL or perilipin. Independently of gender and age, the functional equilibrium between the effects induced by β- and α₂A-adrenergic receptors is correlated with fat-mass distribution and adipocyte hypertrophy. Hypertrophied adipocytes are characterized by a high density of α₂A-adrenergic receptors and a low density of β₁/₂-adrenergic receptors compared with small-sized adipocytes in VAT. Because hypertrophied adipocytes have a much weaker response than visceral adipocytes, lipolysis may be able to prevent untimely FFA release from abundant subcutaneous deposits. These differences explain why VAT, the adipocytes of which have strong lipolytic activity, delivers excess FFA to the liver and perturbs hepatic functions.

Physiological studies confirm most of the results obtained with isolated adipocytes. Lipid mobilization induced by physical exercise is markedly reduced in abdominal SCAT in the obese. Physiological stimulation of α₂A-adrenergic receptors is responsible for defective lipid mobilization [41]. Given such lipolytic defects, it is probable that the recently discovered lipolytic pathway involving the natriuretic peptides is also altered in the obese; however, this requires further study [42]. Increased VAT-derived FFA in the portal vein is correlated with VAT distribution. However, the relative quantity of FFA of visceral origin is lower than the total quantity of FFA originating at other SCAT sites [43,44]. VAT, even if it plays a role in the induction of hepatic insulin resistance, is perhaps not a major determinant factor in the pathogenesis of skeletal-muscle insulin resistance, as it represents only a small percentage of the total systemic FFA delivered by the muscles. VAT adipocytes, characterized by their strong lipolytic reactivity and lower sensitivity to the antilipolytic effects of insulin, undergo more rapid turnover of their stored lipids and represent the FFA-production site responsible for inducing deleterious effects, particularly in the liver [45].

5. AT synthesis of cytokines, hormones and proinflammatory molecules

In addition to its essential functions of lipid storage and FFA management by buffering effects [46], AT has acquired the status of an endocrine organ [47]. Certain AT processes require adipocytes, while others are based on SVF cells. The various cell types present in SVF play important roles in AT secretory activity [21,22,48]. By their secretions, macrophages infiltrating the AT in obese subjects probably affect the function of adipocytes and other cells in the SVF [9,11,17].

Adipocytes produce a substantial quantity of bioactive molecules (lipids and proteins). Among the protein factors, the most studied are leptin and adiponectin (Acrp30 and adipQ), respectively, encoded by the [adipose most-abundant gene transcript-1] apM1 gene) and, more recently, retinol-binding protein-4 (RBP-4) [49,50]. Various factors of the alternative complement system (adipsin, and factors C3, B and D) implicated in ASP and acute-phase protein syntheses (haptoglobin, serum amyloid-A, pentraxin-3, lipocalin 24p3, α₁-acid glycoprotein and cathepsin S) are synthesized by adipocytes. PAI-1, an important factor in the regulation of fibrinolysis found in AT, is not directly derived from adipocytes [51]. Other factors also do not, strictly speaking, originate in adipocytes: inflammatory cytokines [TNF-α, IL-1β, IL-6, IL-8, IL-10, IL-18, transforming growth factor-β (TGF-β) and interferon-γ (IFN-γ)] and chemokines [MCP-1, macrophage inflammatory protein-1α (MIP-1α) and IL-1 receptor antagonist (IL-1Ra)] are thought to act primarily at the paracrine level. Systematic comparative studies of the production capabilities of the cell types found in AT deposits have not yet been undertaken. The various factors secreted by adipocytes and SVF cells, and their primary roles, are summarized in (Table 1), while (Table 2) presents the major differences between VAT and SCAT.

5.1. Leptin

Leptin is a well-known adipocyte hormone. During fasting, low leptin levels stimulate feeding, reduce energy expenditure, and modulate immune and neuroendocrine function to preserve energy stores. On the other hand, rising leptin levels in the overfed state prevent weight gain by inhibiting food intake and increasing energy expenditure. It controls food intake and energy expenditure through its hypothalamic effects. It also exerts multiple peripheral effects. Leptin mRNA expression is lower in VAT adipocytes than in SCAT. VAT adipocytes produce less leptin (per cell and per gramme), independently of cell size, in obese men and women. As stimulation of β-adrenergic receptors negatively regulates leptin release, it may be that any activation of the sympathetic nervous system would preferentially inhibit leptin synthesis by visceral adipocytes, the cells that are the most reactive to physiological amines. Given its mass, its capacity to synthesize leptin and adrenergic regulation of that hormone, VAT contributes modestly to circulating leptin. Only leptin produced by SCAT is well correlated with circulating leptin concentrations.

5.2. Adiponectin

This adipocyte hormone, believed to play an important role in the pathogenesis of insulin resistance, has multiple effects on skeletal muscle, the liver and vessels [52]. Adiponectin possesses antidiabetic and anti-inflammatory properties, and

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Table 1

Adipose tissue: some of the productions/secretions of adipocytes and other cells of the stromal – vascular fraction, grouped according to their contributions to major functions control

| Lipid and lipoprotein metabolism |  |
|---------------------------------|  |
| Lipoprotein lipase (LPL)        |  |
| Acyl-stimulating protein (ASP/C3desArg) |  |
| Prostaglandin E2 (PGE2), prostaglandins F2a (PGF2a) and prostacyclin (PGI2) |  |
| Autotaxin (lysophospholipase D) and lysophosphatic acid (LPA) |  |
| Retinol-binding protein-4 (RBP-4) |  |
| Cholesterol ester transport protein (CETP) |  |

Table 2

Differences in expression/secretion of factors in visceral versus subcutaneous human adipocytes

<table>
<thead>
<tr>
<th>Effects and factors</th>
<th>Regional differences</th>
</tr>
</thead>
<tbody>
<tr>
<td>Miscelaneous secreted factors and hormone receptors</td>
<td></td>
</tr>
<tr>
<td>Leptin mRNA and protein secretion</td>
<td>VAT &gt; SCAT</td>
</tr>
<tr>
<td>Plasminogen activator inhibitor-1 (PAI-1)</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Type-1 11β-hydroxysteroid dehydrogenase activity (cortisone → cortisol)</td>
<td>VAT &gt; SCAT</td>
</tr>
<tr>
<td>Adiponectin (ACRP30)</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Growth-related oncogene factor-α (GRO-α)</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Regulated upon activation, normal T-cell-expressed and -secreted (RANTES)</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Inhibitor of apoptosis cIAP2 mRNA</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Macrophage inflammatory protein-1β (MIP-1β)</td>
<td>VAT &gt; SCAT</td>
</tr>
<tr>
<td>Interleukin-7 (IL-7)</td>
<td>VAT &gt; SCAT</td>
</tr>
<tr>
<td>Tissue inhibitor of metalloproteinases-1 (TIMP-1)</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Thrombopoietin</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Peroxisome proliferator-activated receptor-γ (PPAR-γ) mRNA</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Tumour necrosis factor-α (TNF-α) secretion</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Interleukin-6 (IL-6) secretion</td>
<td>VAT &gt; SCAT</td>
</tr>
<tr>
<td>Omentin</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Insulin-like growth factor-1 and IGF-binding protein-3</td>
<td>SCAT &gt; VAT</td>
</tr>
<tr>
<td>Angiotensinogen</td>
<td>SCAT &gt; VAT</td>
</tr>
</tbody>
</table>

VAT: visceral adipose tissue; SCAT: subcutaneous adipose tissue; NB: some of the differences listed here may increase or reduce according to the extent of the fat mass and adipocyte hypertrophy; also, gender-related differences have not been detailed.

In the few studies comparing adiponectin expression in different fat deposits, the results were inconsistent and often based only on mRNA levels. Two groups, working on tissue explants, found that adiponectin mRNA expression was weaker in VAT than in SCAT. In contrast, no such difference was observed in obese subjects, who produce less adiponectin regardless of fat deposit location [53,54]. A study of isolated adipocytes demonstrated that adiponectin expression and secretion were higher in omental than in subcutaneous adipocytes. Insulin and thiazolidinediones increased its release by omental adipocytes, whereas subcutaneous adipocytes were unaffected [55]. Stimulation of β-adrenergic receptors negatively regulates adiponectin expression (mRNA) and release in VAT. As for leptin, it may be that any sympathetic-nervous-system activation would preferentially inhibit adiponectin production by visceral adipocytes due to their greater reactivity to physiological amines.

5.3. **TNF-α**

The level of synthesis of this inflammatory cytokine by human adipocytes remains highly controversial. Even though its mRNA has been found in those cells, TNF-α is not detected in the veins draining SCAT [56]. The authors of several investigations reported no differences in TNF-α expression between VAT and SCAT. In fact, TNF-α is essentially secreted by SVF cells [48,57]. No notable interfat-deposit differences are evident in the production of this cytokine although, so far, its study has been limited. Macrophages, accumulated in VAT during the development of the fat mass, are capable of synthesizing TNF-α and other cytokines; their functional phenotype is not yet clar-
ified, and noticeable discrepancies are found between results from rodents and humans [58].

5.4. Interleukins-6 and -8

IL-6 and IL-8 are produced by AT. IL-6 is secreted by SCAT and found in the veins draining it [56]. Its synthesis increases in SCAT with BMI and declines with weight loss. VAT produces more IL-6 than SCAT in the morbidly obese [59]. Mammary adipocytes (derived from the differentiation of preadipocytes in vitro) release IL-6, and express its receptor and the signal-transduction protein gp130 [60]. Administration of β-adrenergic agonist increases IL-6 expression in adipocytes as well as in serum [61]. IL-6 produced by VAT and delivered directly to the liver via the portal vein could contribute to the onset of the metabolic syndrome. IL-6 levels are elevated in the portal vein in obese patients [62]. Studies in rodents showed that IL-6 increased the production of very low-density lipoproteins and enhanced the hepatic synthesis of acute-phase proteins. VAT expresses and secretes more IL-8 than does SCAT [63]. It should be noted that most AT-derived IL-6 and IL-8 were probably not made by adipocytes, which have a limited production capacity. SVF cells play a more important role [64].

5.5. IL-1 receptor antagonist (IL-1Ra)

This antagonist is strongly expressed in human AT. IL-1 – which has two forms, IL-1α and IL-1β – is a major mediator of inflammation that acts via a plasma-membrane receptor. Its receptor antagonist, IL-1Ra, is a member of the interleukin family secreted by cells. It is a natural antagonist that binds to the IL receptor, thereby preventing the effects of IL-1α and IL-1β on their target cells. Knockout of the gene encoding IL-1Ra in mice leads to defective lipid accumulation and modified insulin secretion. The anti-inflammatory cytokine IFN-β and leptin stimulate IL-1Ra production in human monocytes and monocyte cell lines. Circulating IL-1Ra levels are markedly elevated in hyperleptinaemic obese subjects. Major weight loss dramatically lowers circulating IL-1Ra levels [65]. IL-1Ra expression is high in human and rodent AT. Explants of human AT secrete IL-1Ra, and lipopolysaccharide (LPS) induces its synthesis [66]. A low-calorie diet leads to substantial lowering of IL-1Ra in AT; macrophages probably play an important role in the processes observed [13].

5.6. Resistin

Resistin is a peptide hormone that comprises 108 amino acids and is expressed in the AT of rodents. Produced during adipogenesis and secreted by rodent adipocytes, resistin is implicated in insulin resistance [67]. It inhibits in vitro adipogenesis and plays a role in the obesity associated with diabetes, at least in rodents. Its role in humans remains uncertain; found in VAT, it is weakly expressed in human adipocytes and originates from macrophages [17]. Resistin expression is strongly induced by the endotoxin LPS and inflammatory cytokines (IL-1, IL-6, TNF-α) in cultured human macrophages [68].

5.7. Visfatin

Visfatin, a recently discovered adipokine that is strongly expressed in AT, has the novel property of stimulating the insulin receptor by binding to a site distinct from that to which insulin binds. It corresponds to a 52-kDa protein expressed in lymphocytes that was identified about a dozen years ago as a B-cell growth factor, called pre-B-cell colony-enhancing factor (PBEF) [69]. Visfatin biology remains poorly understood. Its administration lowers plasma glucose levels in mice [70]. It appears to be preferentially made by VAT. But what are the factors involved in the regulation of its secretion by adipocytes or other visfatin-producing cells in the SVF? The results of a recent study contradicted earlier observations by finding no differences between visfatin mRNA levels in VAT versus SCAT. In addition, there appears to be no relationship between plasma visfatin levels and parameters of insulin sensitivity in humans [71]. Based on the information currently available on visfatin/PBEF, our insufficient knowledge of this new protein secreted by adipocytes and other tissues does not allow any definitive conclusion to be drawn as to its role in the modulation of the effects of insulin and obesity [72].

5.8. Plasminogen activator inhibitor-1 (PAI-1)

PAI-1, a physiological inhibitor of plasminogen activation, plays an essential role in the control of the fibrinolytic system. PAI-1 (mRNA) expression, measured in cultured AT from nonobese subjects, appears to be higher in VAT. In contrast, the results of a study of obese individuals showed excess PAI-1 mRNA and enhanced PAI-1 secretion in SCAT. PAI-1 is synthesized in endothelial cells, and a non-negligible part of AT PAI-1 may be of nonadipocyte origin. PAI-1 expression in VAT could be associated with stromal cell density [51]. Visceral and omental adipocyte production of PAI-1 have also been confirmed. IL-6 or oncostatin M, proinflammatory mediators, selective ligands of cytokine-receptor proteins and gp130 stimulate PAI-1 synthesis in VAT in vitro explants, and in human preadipocytes and adipocytes [73]. Macrophages that accumulate in the AT of obese subjects may also contribute, via oncostatin production, to the enhanced PAI-1 synthesis observed in the obese.

5.9. Fibroblast growth factors (FGF)

FGF are proteins that bind to heparin, and play important roles in development and organogenesis. Injected FGF2 binds to basement-membrane proteins and can induce AT development at the injection site [74]. Enhanced FGF2 immunoreactivity was detected in the VAT of obese subjects. FGF9 was also found in human AT. These observations suggest that, if FGF were to be locally produced in AT, they would affect adipogenesis and angiogenesis, and modulate AT growth and distribution.

5.10. Vascular endothelial growth factor-A (VEGF-A)

Vascular endothelial growth factor-A is a major regulator of angiogenesis and vessel-wall permeability. This factor is
induced by hypoxia in endothelial and smooth muscle cells, and in human adipocytes [75]. It is expressed and secreted by murine AT, preadipocytes and adipocytes. Agents that stimulate protein kinase-A or -C increase VEGF-A expression in rodent preadipocytes [76]. Inflammatory cytokines, such as IL-6 and oncostatin, stimulate VEGF-A production and angiogenesis in VAT and SCAT [77]. It is released by human adipocytes, although the majority of VEGF-A released by cultured AT explants appears to originate from SVF cells [48]. A β-adrenergic agonist has no influence on VEGF-A production by human adipocytes, but insulin does stimulate its release. This effect suggests that elevated insulin levels, in addition to their impact on TG synthesis, might trigger angiogenesis in AT [78].

5.12. Sex hormones

Cytochrome P-450 aromatase (P-450arom), produced by the CYP19 gene, is expressed in AT stromal cells. It is implicated in the local production of oestrogens [84] and is especially expressed in postmenopausal women. The transcript level is higher in gluteal than abdominal SCAT. The oestrogens synthesized most probably exert local paracrine or intracrine actions. Adipocytes are sensitive to oestrogens, although the data for the levels of oestrogen receptors (ER-α and ER-β) and their transcriptional regulation have been mixed.

Sex steroids have multiple effects on the distribution of fat mass, and on the elements controlling lipolytic pathways in men and women. Women suffering from polycystic ovary syndrome (PCOS), characterized by a hyperandrogenic state, are prone to abdominal obesity, whereas testosterone-treated men have fewer fat deposits, with selective loss of VAT rather than SCAT. Testosterone leads to lower lipolytic activity in SCAT with no effects on VAT response, attributed to decreased expression of HSL and β2-adrenergic receptors [85]. Oestrogen regulates α2-adrenergic-receptor expression and attenuates the lipolytic response in SCAT, but not in VAT [86].

5.13. Cortisol and 11β-hydroxysteroid dehydrogenase-1

Excess cortisol is associated with VAT accumulation and regression of peripheral fat. This distribution pattern was attributed to the relative density of glucocorticoid receptors in fat deposits, with increased density in omental AT. The response to exogenous corticosteroids is more intense in omental AT [87]. Cortisol is synthesized locally in AT from inactive cortisone. 11β-hydroxysteroid dehydrogenase-I (11β-HSD1) activity has been characterized in AT stromal cells and adipocytes; it is higher in VAT than in SCAT [88]. Although the association between excess cortisol (of endocrine or paracrine origin) and visceral obesity has been clearly established in rodents, the true impact of 11β-HSD1 expression and activity in obese humans remains to be clarified [89]. 11β-HSD1 activity is increased in the VAT of obese men [90], and the 11β-HSD1 gene expression is enhanced in the SCAT of obese women with metabolic syndrome [91]. VAT is most probably the production site of the cortisol found in the splanchnic circulation [92].

6. Conclusion and future trends

There is no doubt that VAT distribution precedes the clinical presentation of the metabolic syndrome (insulin resistance, glucose intolerance, hypertension and dyslipidemia) that will progress to associated pathologies (T2D, cardiovascular disease) without appropriate interventions. The determinative role of visceral obesity in the metabolic syndrome is accepted by the majority of endocrinology, diabetology and cardiology societies. However, a gray area persists in our understanding of the relative influences of metabolic (associated with the hepatic effects of VAT-derived FFA) and humoral (adipokine and cytokine synthesized by VAT) contributions to the abnormalities observed. Indeed, to date, no study results have refuted the role of VAT-derived FFA in the pathogenesis of hepatic insulin resistance; the mechanisms have been clearly established and are discussed above. Studies conducted on rodents and dogs fed high-fat diets [45] confirm the important role of FFA in the induction of hepatic insulin resistance. However, VAT is not the only source of systemic FFA; depending on the respective degrees of VAT and SCAT distribution, FFA delivered by nonvisceral abdominal AT cannot be dismissed in the development of insulin resistance, especially when it affects skeletal muscle [44].

Adipokines and cytokines produced by adipocytes or SVF cells are thought to play major roles in pathological states such as morbid obesity and T2D [15]. Nevertheless, the precise metabolic role of the most recently discovered adipokines such as resistin, visfatin, RBP-4 and SAA remains poorly understood, and the subject remains controversial. The results summarized in this review have delineated the secretion characteristics of VAT compared with those of SCAT. This information was derived mainly from in vitro experiments that provided no information on the bioavailability of the molecules produced. In addition, the lack of homogeneity across observations must be borne in mind. The discrepancies could be due to the poor characterization of the cell populations evaluated or to the heterogeneous distribution of AT deposits, reflecting the patients examined and the different experimental methods used in the comparative studies. At present, the products that are derived from VAT and gain access to the liver via the portal vein need to be fully characterized before the true impact of FFA and the adipokines can be ascertained.

One of the novel developments of research into obesity has been the characterization of a low-grade chronic inflammatory state that worsens with the patient’s weight gain. That an inflam-
imentary response occurs in obesity is now well accepted [93]. It appears to be triggered in adipose tissues; the liver, in particular, may also be involved as the disease becomes established. Because AT (adipocytes and SVF cells) expresses and secretes numerous proteins linked to inflammatory processes, it may be that adipokines and other molecules with inflammatory properties originating in AT contribute to the chronic activation of the immune system in the obese. The contribution of the liver is also important and sometimes overlooked. The mechanisms that lead to AT colonization by macrophages in the obese as well as the role played by AT macrophages remain to be elucidated. Assessing the possible impact of the process on the progressive loss of insulin sensitivity and the development of the metabolic syndrome requires further in vitro studies and clinical investigations.

References


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