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How does adipose tissue contribute to inflammageing?

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ABSTRACT

Across aging, white adipose tissue (WAT) undergoes significant changes in quantity and distribution, with an increase in visceral adipose tissue, ectopic fat deposition and a decline in gluteofemoral subcutaneous depot.

In particular, WAT becomes dysfunctional with an increase in production of inflammatory peptides and a decline of those with anti-inflammatory activity and infiltration of inflammatory cells. Moreover, dysfunction of WAT is characterized by preadipocyte differentiation decline, increased oxidative stress and mitochondrial dysfunction, reduction in vascularization and hypoxia, increased fibrosis and senescent cell accumulation.

WAT changes represent an important hallmark of the aging process and may be responsible for the systemic pro-inflammatory state ("inflammageing") typical of aging itself, leading to age-related metabolic alterations.

This review focuses on mechanisms linking age-related WAT changes to inflammageing.

1. Introduction

Aging is the main risk factor for multiple chronic diseases, decline in physical function and frailty. Such conditions are frequently preceded by metabolic alterations and low-grade chronic inflammation. Adipose tissues are organized to form a large adipose organ with discrete anatomy, vasculature and innervation, high plasticity and complex cytology where the main cells, the adipocytes, are defined as white or brown in relation to their different morphology and function (Cinti, 2012). Across aging, both white adipose tissue adipose (WAT) and brown adipose tissue (BAT) undergo changes in quantity, distribution and function. WAT changes represent an important hallmark of the aging process itself and may be responsible for the systemic pro-inflammatory state typical of aging itself (Kirkland et al., 2002). This age-related state of chronic sterile low-grade inflammation has been named "inflammageing" and participate in the development of frailty, disability and most chronic degenerative diseases including age-related cardiovascular and cerebrovascular diseases (Franceschi, 2007; Franceschi and Campisi, 2014). Even if a variety of tissues and organs participate in inflammageing (Franceschi and Campisi, 2014; Franceschi et al., 2018; Liberale et al., 2020), WAT plays a major role. Moreover, WAT plays a major role in human physiology by constituting the main reservoir of energy stores, deposited as triglycerides and by serving as an endocrine organ secreting several peptides which have a fundamental role in the regulation of energy homeostasis, insulin sensitivity and cardiovascular function (Cinti et al., 2005; Giordano et al., 2013). WAT is usually divided into visceral (VAT) and subcutaneous (SAT) adipose tissue (Bosello and Zamboni, 2000). SAT is where the majority of excess lipids are stored, and is considered healthier than VAT in both lean and obese subjects while, VAT, being more metabolically active and sensitive to lipolytic stimulus, is strongly related to the onset of metabolic syndrome and to increased cardiovascular morbidity and mortality (Bosello and Zamboni, 2000).

Therefore, the aim of this review is to revise current literature about the contribution of WAT to inflammageing and explore potential mechanisms at the systemic, tissutal and cellular levels.

2. Age related body fat redistribution

As people age, a progressive increase in fat mass occurs, in response

Abbreviations: WAT, white adipose tissue; BAT, brown adipose tissue; VAT, visceral adipose tissue; SAT, subcutaneous adipose tissue; FFA, free fatty acids; PPAR-gamma, peroxisome proliferators-activated receptor gamma; C/EBP-alpha, CCAAT/enhancer-binding protein alpha; NF-κB, nuclear factor kappa B; TNF-alpha, tumor necrosis factor alpha; IL-6, interleukin-6; MCP-1, monocyte chemoattractant protein-1; ROS, reactive oxygen species; HIF-1-alpha, hypoxic factor 1 alpha; SASP, Senescence Associated Secretory Phenotype; NKT, natural killer cells; TGF-beta, transforming growth factor beta; ER, endoplasmic reticulum; ECM, extra cellular matrix; ERK, extracellular signal-regulated kinase.

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to chronic positive calorie balance, reduced physical activity and lower basal metabolic rate (Mancuso and Bouchard, 2019). An increase in total adiposity may be independent of body weight changes due to a concomitant decrease in muscle mass observed with aging, so-called sarcopenia (Prentice and Jebb, 2001), as well as a decrement in total body water, especially the intracellular compartment, and a decreasing trend of bone mass (Buffa et al., 2011).

Moreover, across aging, progressive redistribution of WAT from the periphery of the body (i.e. loss of gluteofemoral SAT) to the abdomen occurs (i.e. increase of VAT) (Zamboni et al., 2005). With aging, accumulation of abdominal fat and deposition of ectopic fat occur independently of weight gain resembling the phenotype identified as TOFI, i.e. thin-on-the-outside fat-on-the-inside, with an increased metabolic risk (Thomas et al., 2012). Age-related dysregulation of lipid metabolism in subcutaneous adipocytes, in particular in the gluteofemoral regions, is associated with triglyceride spillover and could in part explain not only the increases in visceral adiposity but also in ectopic fat deposition within non-adipose tissues such as the skeletal and cardiac muscles, liver, pancreas, kidney and vessels (Rossi et al., 2011) (Fig. 1). Increased free fatty acids (FFA) delivery to other organs, especially when combined with defective FFA oxidation, leads to the accumulation of "toxic" lipids such as ceramides and diacylglycerols, thereby causing lipotoxicity (Unger, 2005), insulin resistance (Mazzali et al., 2006; Zoico et al., 2010b), hemodynamic age-related alterations (Zamboni et al., 2014) and organ dysfunction (Tardif et al., 2014).

Interestingly, obesity and weight gain, along with aging, are strongly related to the increase of VAT and to ectopic fat deposition (Zamboni et al., 2014). Moreover, both obesity and weight gain may also speed up the onset of age-associated diseases, which further emphasizes the impact of WAT in aging (Zamboni et al., 2014).

Aging is characterized not only by an increase in WAT, but also by a decline in BAT (Cinti, 2012; Saely et al., 2012). The main function of brown adipocytes is the dissipation of energy through uncoupled respiration to produce heat; this mechanism is mediated by a carrier protein, noted as thermogenic factor uncoupling protein-1 (UCP-1) expressed on the inner membrane of mitochondria (Cinti, 2012). There are many possible mechanisms that link aging to BAT involution, such as the loss of mitochondrial function, impairment in the sympathetic nervous system, age-induced alteration of brown adipogenic stem/progenitor cell function, and changes in endocrine signals (Zoico et al., 2019b).

Aging is also associated with a reduction in beige adipocyte formation (Cinti, 2012). Beige adipocytes differentiate from a subpopulation of WAT-resident progenitors; a defective ability of progenitors cells to proliferate and differentiate has been hypothesized with aging, probably due to changes in trophic factors in the adipose tissue microenvironment (Zoico et al., 2019b). Recently, microbiota changes with aging have also been suggested as another potential mechanism that may influence browning of WAT (Moreno-Navarrete and Fernandez-Real, 2019).

3. Age related WAT dysfunction

WAT is organized in a large adipose organ with discrete anatomy, vasculature and innervation, specific cytology and high plasticity (Cinti, 2012). WAT is composed of different cell types including the adipocyte fraction, which contains primarily mature adipocytes and the stromal

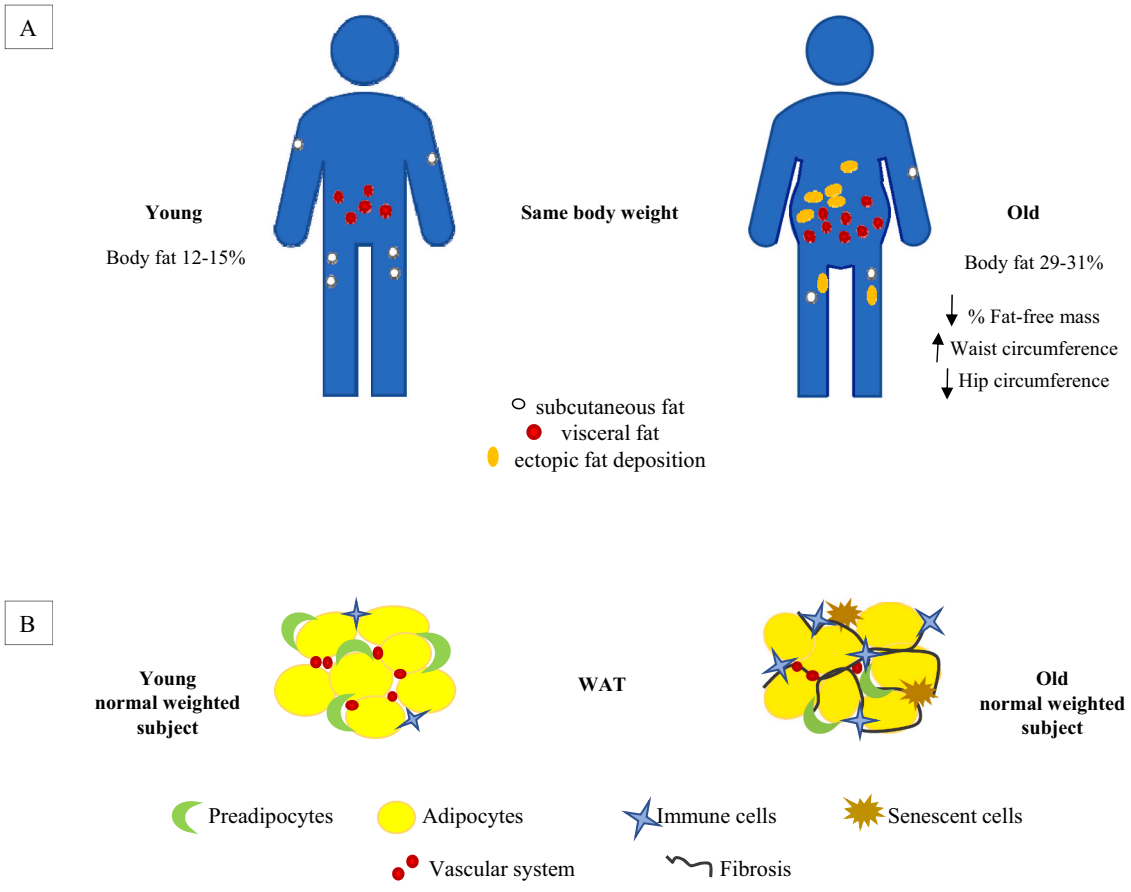

Fig. 1. A) Body composition in young and old normal weighted individuals with the same BMI. Subcutaneous fat (white), visceral fat (red) and ectopic fat deposition (yellow) in muscle, heart, liver.
B) White adipose tissue (WAT) characteristics in young and old normal weighted individuals with the same BMI. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

fraction, composed of adipogenic stem cells, preadipocytes, macrophages, lymphocytes, endothelial cells, pericytes, and fibroblasts (Cinti et al., 2005; Cinti, 2012; Cawthorn et al., 2012; Senesi et al., 2019). With aging WAT undergoes significant morphological and functional changes which lead to its dysfunction, in particular to preadipocyte differentiation decline, inflammation, morphological changes and senescent cell accumulation (Fig. 2).

3.1. Preadipocyte differentiation decline

Adipocytes derive from progenitors cells called preadipocytes, which represent 15–50% of cells in WAT and play an important role in WAT maintenance and regeneration.

Adipogenic differentiation is characterized by morphological changes, lipid accumulation and expression of genes typical of fat cells; this process is under the control of a transcriptional cascade involving specific adipogenic key transcriptional factors. Several studies, conducted both in animals and humans, showed that preadipocytes reduce their ability to replicate and differentiate, and thus to store lipids with aging (Sepe et al., 2011).

Several observations support the decline of preadipocyte differentiation with aging. Caso et al. (2013), observed that the rate of replication and differentiation of preadipocytes isolated from subcutaneous peripheral fat in old subjects was significantly lower than that of healthy young individuals. Schipper et al. (2008) isolated human preadipocytes from different subcutaneous fat depots of 12 female donors and observed a decrease in both their replicative potential, and their differentiation capacity with increasing donor age. They also observed that serial in vitro passages of preadipocytes diminished their ability to differentiate into mature adipocytes with increased expression of SA-β-gal, caveolin-1, as well as of senescence markers (Schipper et al., 2008). Furthermore, when comparing preadipocytes in healthy young, middle-aged, and aged volunteers it was observed that aged preadipocytes had a higher expression of senescence markers, antiapoptotic genes, proapoptotic gene BAX, upregulation of NF-κB (nuclear factor kappa B) and a downregulation of genes associated with tissue renewal capacity such as p53, caspase 3, caspase 8, and caspase 9 (Alt et al., 2012).

Age-related decline in pre-adipocyte differentiation has been shown to be linked to a decreased expression of the adipogenic transcription factors CCAAT/enhancer-binding protein (C/EBP)-alpha and of peroxisome proliferators-activated receptor gamma (PPAR-gamma) (Stout et al., 2016). Several factors may affect the activity of these transcription factors. Tumor necrosis factor alpha (TNF-alpha), secreted at a considerably higher amount in WAT of older subjects (Lumeng et al., 2011), has been found to have a key role in the age- related decline of preadipocyte differentiation (Pararasa et al., 2015). This has been clearly shown by Tchkonia et al. (2007) who demonstrated that in preadipocyte culture derived from pathogen-free male Brown Norway rats, TNF-alpha decreases the expression of C/EBP-alpha and PPAR-gamma and induces some inhibitors of adipogenesis such as CHOP, CUGBP and C/EBP-b-LIP. Furthermore, in 3T3-L1 preadipocytes, inhibition of TNF-alpha through adiponectin increased preadipocyte differentiation and cell viability, thereby upregulating PPAR-gamma and C/EBP-alpha (Yang et al., 2018).

It is known that WAT presents an infiltration of inflammatory cells such as macrophages, B and T lymphocytes and neutrophils, (Cox et al., 2019; Lumeng et al., 2011; Pyrina et al., 2020) which may also be linked to a decline in preadipocyte differentiation. Macrophages, through the production of nitric oxid, may increase the level of the hypoxia-inducible-factor 1 alpha (HIF-1 aplha), determine DNA damage and p53 phosphorylation, as well as decrease mitochondrial biogenesis in preadipocytes (Jang et al., 2016).

The ability of preadipocytes to differentiate can be influenced also by both chronic and intermittent oxidative stress (Findeisen et al., 2011; Houstis et al., 2006). In fact, increased oxidative stress decreases preadipocyte differentiation through the inhibition of different steps in the cell cycle, such as the expression of S-phase genes downstream of the retinoblastoma protein (Findeisen et al., 2011). Moreover chronic oxidative stress, obtained in vitro by intermittent hypoxia, may determine decreased mRNA levels of of C/EBP-alpha and PPAR-gamma in preadipocytes (Fernando et al., 2020).

Intermittent hypoxia has been also shown to be toxic for the preadipocytes and causes increased generation of mitochondrial ROS, increased prevalence of cells with nuclear localization of p16 and higher prevalence of cells positive for senescence-associated beta-galactosidase (Polonis et al., 2020).

Interestingly, oxidative stress and hypoxia, besides decreasing preadipocyte differentiation into adipocyte, may also induce the preadipocyte to acquire surface markers such as F4/80, CD80 and CD 86 and differentiate into macrophages (Engin, 2017; Zhang et al., 2018).

With aging, dysfunction of preadipocytes could compromise their ability to store lipids thereby increasing the exposition of WAT to oxidative stress (Sepe et al., 2011). The abundance of FFA induces a

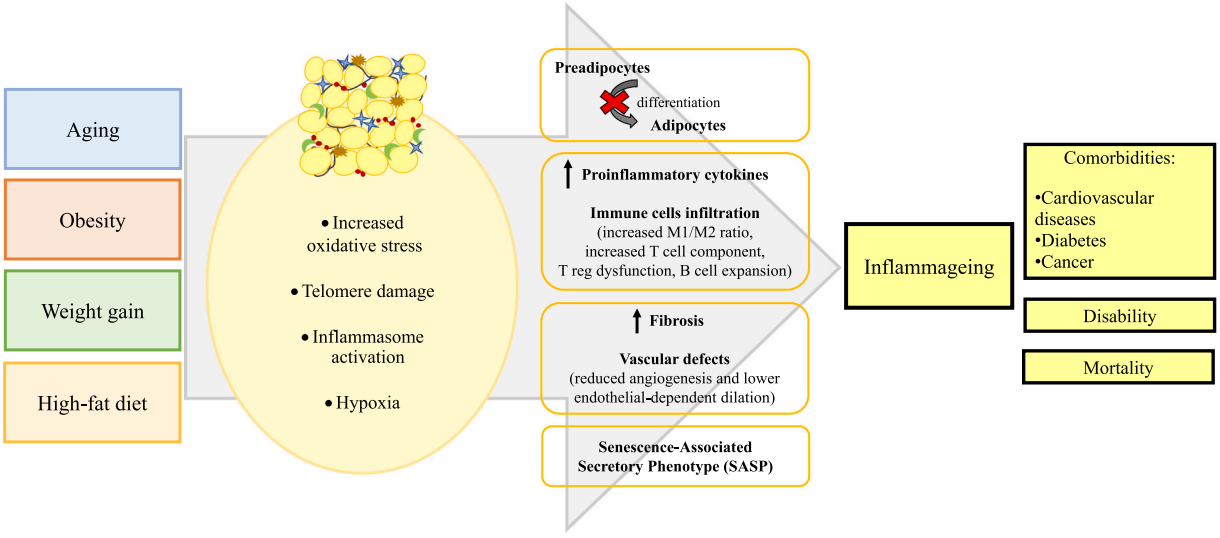

Fig. 2. Main aspects of the aging white adipose tissue (WAT): morphological and functional characteristics together driving to WAT dysfunction and clinical consequences.

reduced expression of the enzymes required for their processing into triglycerides (Guo et al., 2007) and their accumulation could increase the release of cytokines from adipose tissue itself (Suganami et al., 2005). In a murine model of aged mice, increased FFA induces pre-adipocytes apoptosis and a decrease in adipogenesis (Guo et al., 2007; Stout et al., 2016).

High-fat diet might also influence fat cell homeostasis, thus increasing hypoxic stress through over-expression of HIF-1-alpha (Jang et al., 2016), as well as through an increase in TNF-alpha (Bloom et al., 2020). High-fat diet could influence the expression of genes involved in the regulation of telomere dynamics in WAT, which might result in increased inflammation (Bloom et al., 2020).

Taken together, these studies strongly support that preadipocytes become dysfunctional with age; moreover, dysfunctional preadipocytes do not differentiate into adipocyte, contributing to lipodystrophy, inflammation, insulin resistance, and metabolic alterations (De Carvalho et al., 2019). However it is still unknown what threshold of pre-adypocyte dysfunction is needed to cause these alterations, and if preadipocytes dysfunction itself is enough to cause such changes and whether preadipocyte dysfunction is prevalent in SAT or VAT with aging.

3.2. Adipokines and aging

Besides being responsible for energy storage, nutrient sensing and temperature regulation WAT is recognized as the largest endocrine and immune organ of the body (Cinti, 2012). Since the discovery of leptin, a variety of adipokines have been shown to be secreted by adipocytes (Cinti, 2012). Adipocyte size is one of the factors regulating cytokines release within adipose tissue, as they become hypertrophic and more inflammatory (Skurk et al., 2007).

In general, with aging, the majority of adipokines are elevated in comparison with younger individuals (Mancuso and Bouchard, 2019). The relation between aging and endocrine function of WAT is complex to study in humans because aging is strictly associated with changes in fat mass and fat distribution as well as with a high prevalence of metabolic syndrome, insulin resistance and obesity. In fact, age-related VAT increase and/or concurrence of obesity and/or metabolic syndrome, may all be responsible for an increase in inflammatory and a decline in anti-inflammatory adipokines production.

In vitro older adipocytes, as opposed to young mature adipocytes, display not only reduced gene expression of adiponectin and leptin, but also a significant increase in proinflammatory cytokine production, such as interleukin-6 (IL-6) and monocyte chemoattractant protein-1 (MCP-1), as well as an increase in insulin resistance (Yu and Zhu, 2004; Zoico et al., 2010a). In vitro aged adipocytes accumulate ROS, increase mRNA expression of key proteins related to the remodeling of the extracellular matrix as well increase p53, p21 and p16 expression, compared to younger cells (Zoico et al., 2019a).

Compared to younger adults, older subjects show higher fasted and postprandial concentrations of leptin (Johnson et al., 2020). Leptin secretion profile has been shown to be different in both young and aged people (Mancuso and Bouchard, 2019). At a young age, leptin is predominantly secreted by SAT, while in older age, leptin is secreted by VAT (Carter et al., 2013). Despite an increase in the plasma level of leptin in old people, there is a disorder and weakness in the performance of leptin in old age (Gabriely et al., 2002). Lower availability of leptin in the hypothalamus, impaired peripheral leptin action, or both, have been proposed as mechanisms of leptin resistance through aging (Scarpace and Tümer, 2001). In humans, an increase in leptin resistance with both adiposity and aging has been observed (Zoico et al., 2008).

In contrast to other adipose-derived adipokines, adiponectin is inversely related to total adiposity and VAT, and shows insulin-sensitizing and anti-atherogenic properties (Mancuso and Bouchard, 2019). In vitro adiponectin has been shown to improve preadipocyte differentiation (Yang et al., 2018). Moreover, in vitro pre-treatment with globular or full-length adiponectin significantly suppressed LPS-induced expression of IL-6 mRNA, MCP-1 mRNA in 3T3-L1 adipocytes, reduced NF-κB activity by 50% and 40% and increased PPARgamma mRNA levels (Zoico et al., 2009).

Serum adiponectin has been shown to increase with aging in humans (Kizer et al., 2010; Kizer et al., 2012) and higher levels of adiponectin have been observed in centenarians (Atzmon et al., 2008; Arai et al., 2019). Despite the basic science supporting the beneficial metabolic and anti-inflammatory effects of adiponectin, its association with morbidity and mortality, at least in patients with cardiovascular disease as well as in the elderly, is still controversial. A negative association between adiponectin and coronary heart disease has been observed in healthy middle-aged cohorts, (Pischon et al., 2004; Pischon et al., 2011), while a positive association with mortality was found in the elderly (Wannamethee et al., 2007; Poehls et al., 2009). Among the participants of the Health ABC Study, a significant association was observed between adiponectin and risk of incident disability and all-cause mortality (Baker et al., 2011), but is an association that is no longer significant after adjusting for weight loss and physical performance at baseline; these findings are in line with the "adiponectin paradox".

As a matter of fact, secretory profile of WAT became more proinflammatory with aging. Leptin resistance is a feature of aging, and the adiponectin paradox seems to occur.

3.3. Sirtuins and WAT

An important role is emerging for sirtuins as anti-inflammatory mechanism that could be reduced with aging, thus representing a potential mechanism involved in inflammageing.

Sirtuins are a family of NAD+-dependent protein deacetylases, with different cellular localization, and are able to deacetylate target proteins, regulate gene expression and modulate transcription factors implicated in many biological processes (Houtkooper et al., 2012, Haigis and Sinclair, 2010). SIRT-1 is ubiquitously expressed and is present in WAT. Overexpression of SIRT-1 in 3T3-L1 cells reduces adipogenesis and triglyceride accumulation; resveratrol, through SIRT-1 activation, promotes lypolisis and reduces fat accumulation (Picard and Guarente, 2005). Moreover SIRT-1 has been shown to suppress NF-κB activity and decrease inflammation through several mechanisms (Mendes et al., 2017). SIRT-1 exerts its anti-inflammatory activity also by reducing oxidative stress through inhibition of iNOS activity and down-regulation of COX-2 expression (Schug and Li, 2011).

SIRT-1 not only has anti-inflammatory properties, but also anti-apoptotic and anti-aging effects (Mendes et al., 2017). In fact, SIRT-1 is involved in cellular senescence and is strongly downregulated in senescent cells (Ota et al., 2007).

Until now and to the best of our knowledge, studies specifically evaluating in WAT the role of sirtuins decline with aging in inflammageing are scarce; they have been prevalently performed in vitro and therefore deserve further investigation.

3.4. WAT immune cell infiltration

The cross-talk between immune cells and WAT has been suggested as an important regulator of WAT function and systemic metabolism (Chung et al., 2018).

Across aging, immune cell infiltration including M1 and M2 macrophages, T and B cells, and granulocytes tends to accumulate in WAT as a feature of dysfunctional aged WAT (Garg et al., 2014).

T cells and M1 macrophages are known to contribute to creating a pro-inflammatory environment, mostly in the VAT compartment (Lumeng et al., 2011), while M2 macrophages and T regulatory cells (T reg) normally release anti-inflammatory cytokines such as interleukin IL-10 and transforming growth factor beta (TGF-beta), which in turn increase insulin sensitivity and inhibit adipose tissue inflammation and dysfunction. Dynamic changes of these immune cells in WAT underpin

their involvement in pathologies associated with WAT dysfunction (Stout et al., 2016; Guzik et al., 2017).

Macrophages were the first immune cells identified in WAT and the most abundant cell type in both VAT and SAT. Macrophages in VAT remain substantially stable in number during aging, but the ratio between pro-inflammatory M1 and anti-inflammatory M2 macrophages increases with aging, due to various drivers (Garg et al., 2014; Antony et al., 2018). In a murine model, a decrease in the resident CD206+ macrophages was found in WAT, modifying the ratio in favor of inflammatory macrophage subtypes (i.e. CD11c+ and CD206- CD11c-) (Trim et al., 2018). Moreover, this imbalance appeared to be associated with a decrease in PPAR-gamma expression (Lumeng et al., 2011; Mancuso and Bouchard, 2019). In addition to these observations, it should be taken into account that, like obese WAT macrophages, aged ones display high endoplasmic reticulum (ER) stress and have been associated with an increased production of pro-inflammatory cytokines by the WAT. An elevated ER stress response was observed in aged WAT stromal cells, which were also more sensitive to ER stress (Ghosh et al., 2016). Elevated ER stress and inflammation of aged WAT have been linked to impaired autophagy, demonstrated by the accumulation of some autophagy substrates in old stromal vascular fraction, such as LC3-II and p62 (Ghosh et al., 2016).

Aged WAT creates also a favorable microenvironment for T and B lymphocyte activation, and lymphocytes constitute the second most abundant immune cell population in VAT. Antony et al. (2018) observed a 2-fold increase in CD3+ T cells in WAT of aged mice, compared to young animals. The greatest increase regarded CD8+, rather than CD4+ T cells (Antony et al., 2018). The key molecules that mediate T cell infiltration into WAT in aging are still not precisely defined. Accumulation of CD8+ T cells in aged WAT, in line with what is observed in obesity, may contribute to increased adipose tissue inflammation (Antony et al., 2018).

Enrichment in T reg cells in WAT of old mice has been observed with a continuous rise from young individuals to old ones. Hence, by suppressing T cell response, T reg dysfunction may be associated with increased susceptibility to alterations such as age-related insulin resistance (Bapat et al., 2015). In fact, when compared to T cells of young mice, T reg of aged mice had a substantial increase in a set of transcripts (PPAR-gamma, Gata-3, Klrg1, Ccr2), which may result in local adaptation to a lipolityc and hypoxic adipose tissue environment (Bapat et al., 2015).

T reg in aging WAT are also known to express high levels of ST2, a receptor for IL-33 (Antony et al., 2018). Recently, a subpopulation of γ/δ T cells, termed PLZF+ γ/δT cells, has been demonstrated to play a considerable role in age-related WAT T reg accumulation via producing IL-17A, which in turn induces stromal cell production of IL-33 (Antony et al., 2018). Interaction of TCR-antigen-MHC II on preadipocytes and cytokines such as IL-33, may promote T reg accumulation in aging (Guzik et al., 2017). γ/δ T cells have been demonstrated to represent a substantial proportion of T cells in the WAT and their number increases with aging and in metabolic and vascular pathologies (Guzik et al., 2017). These cells are also an important source of strongly pro-inflammatory IL-17 (Guzik et al., 2017).

Furthermore, IL-6 and IL-17 are regulators of the recruitment of granulocytes and CD11b+cells, promoting also ectopic fat accumulation (Guzik et al., 2017). This process is in part controlled by decreased microRNA expression such as miR26a, providing also a link to cardiac injury (Guzik et al., 2017). Finally, T cell presence and activation in dysfunctional aged adipose tissue is also closely linked to inflammasome activation (Guzik et al., 2017). Nlrp3 regulates IL-18 and IFN-γ in WAT and promotes effector T cell accumulation (Chimin et al., 2017).

With aging, B-cell content also increases in WAT, promoting activation of other immune cells and affecting metabolic status (Guzik et al., 2017; Trim et al., 2018). B cells have been shown to accumulate in aged WAT within the fat-associated lymphoid clusters (Camell and Dixit, 2019) and produce proinflammatory cytokines mediating WAT inflammageing and age-associated insulin resistance (Khan et al., 2019).

On the other hand, a significant decrease with aging is observed in WAT invariant natural killer cells (NKT), a type of lipid-sensing innate T cell, which may assist in regulating T reg number and function in young mice by producing IL-2 (Antony et al., 2018; Wang et al., 2019).

Finally, mast cells number also increases in dysfunctional aged WAT and this increase has also been linked to atherosclerosis and metabolic dysfunction by promoting monocyte recruitment (Guzik et al., 2017); in contrast, eosinophil content in WAT shows no or only modest changes in aging (Guzik et al., 2017).

In definitive, a great variety of inflammatory cells infiltrate with aging into WAT. More detailed knowledge of WAT immune cell population infiltration with aging could be fundamental in order to identify a pathophysiological basis responsible for age-associated metabolic alteration, inflammageing and chronic diseases.

3.5. *WAT morphological changes*

Adipose tissue is composed of several cell types including not only adipocytes and their precursor cells, but also endothelial cells, pericytes, fibroblasts and immune cells, which are organized in a complex architecture (Coelho et al., 2013; Tchkonia et al., 2010).

Some morphological changes occur in WAT with aging. Advanced age has been associated with an increased presence of small dysfunctional 'mesenchymal adipocyte-like' cells (Kirkland and Dobson, 1997) with a reduced capacity for lipid accumulation. Larger lipid droplets form in preadipocytes of young animals compared to those from old animals after a high fat diet (Guo et al., 2007). In this study a leftward shift was observed in the adipocyte area histogram, with approximately 70% of all adipocytes from VAT of the old mice falling into the two smallest adipocyte areas, compared to only approximately 10–15% of the adipocytes from young samples (Guo et al., 2007). After 10 weeks of a high fat diet, adipocyte diameters were significantly lower in old rats (25-month old) than in young rats (6-month old), suggesting age-related adipocyte incompetence, i.e. a failure in FFA accumulation (Tardif et al., 2014). On the other hand, an increase in adipocyte diameter has been found in vitro with the aging of the cell from post-induction day 10 to 26 (Zoico et al., 2019a). To the best of our knowledge, no study compares adipocyte diameter in humans in VAT and SAT between young and old subjects, and it is also unknown, what the critical size of adipocytes could be in healthy aging.

Greater WAT fibrosis has been observed in the WAT of aged mice (Donato et al., 2014). A murine model of aged mice lacking the relaxin receptor RXFP1 showed an increase in collagen deposition in WAT, with both pericellular deposition and accumulation of thick collagen fibrils (Bennett et al., 2019). The molecular mechanisms that direct adipose cell conversion towards a pro-fibrotic phenotype are not completely understood. It has been shown that progenitor cell activation by TGF-beta/Smad signaling plays a role in combination with inflammatory cytokines secreted by inflammatory cells (Sun et al., 2013). Macrophages might be involved in the increased fibrosis of WAT with aging through the induction of mitochondrial dysfunction in preadipocytes, with a consequent increase in HIF-1a and activation of pro-fibrogenic pathways (Jang et al., 2016). In an in vitro model of adipocyte aging, we have recently found that aging adipocytes are associated with an alteration of ECM and fibrosis, with increased production of collagen VI-A3, paralleled by a decrease in caveolin-1 expression and a modulation of the ERK pathway (Zoico et al., 2020).

The increased fibrosis seen in aged WAT is usually concomitant with, and may be responsible for, vascular dysfunction. WAT is a relatively hypoxic tissue that receives about 5% of total cardiac output in young adults (Blogowski et al., 2012). Hypoxia in WAT increases with weight gain, obesity (Pasarica et al., 2009) and aging (Donato et al., 2014; Zhang et al., 2011). This has been observed in animals and in vitro. Donato et al. (2014) compared measures of VAT artery function in young (6.1 ± 0.4 months) and old (29.6 ± 0.2 months) B6D2F1 mice and

observed that aging determined a reduction in vascularity, as well as in angiogenic capacity and in the expression of vascular endothelial growth factor in adipose tissue. Finally, they also found that endothelium-dependent dilation was lower in isolated arteries from VAT of the old mice, suggesting that, taken together, these vascular defects contribute to the increase in oxidative stress and might be a potential contributor to tissue hypoxia itself (Donato et al., 2014). The hypoxia rate of adipose tissue was found to be increased with advanced age in both SAT and VAT of aged C571/6 mice (Zhang et al., 2011). Furthermore, exposure of 3T3-L1 adipocytes to the levels of hypoxia, as observed in aging adipose tissue, has been shown to alter multiple aspects of adipose biology, inducing increased levels of insulin stimulated glucose uptake and decreased lipid content.

Morphological changes observed with aging in WAT, and in particular the increase in fibrosis and the decline in vessels, may be the consequence of a greater inflammation, and may concur to its dysfunction.

3.6. Telomer shortening and senescent cell accumulation

Telomere shortening is a known mechanism of premature cellular senescence (Zhu et al., 2019). Besides aging, both insulin resistance (Gardner et al., 2005; Tzanetakou et al., 2012) and inflammation (Minamino et al., 2009) have been shown to be associated with an increased expression of telomerase enzyme activity, and thus shorter telomeres. Dysfunctional WAT telomeres trigger a p53-dependent response (Gardner et al., 2005; Tzanetakou et al., 2012) which could modify adipocyte metabolism that leads to WAT inflammation, dysfunction and insulin resistance (Vergoni et al., 2016). Shorter telomeres also promote infiltration of macrophages into adipose tissue (Minamino et al., 2009) and might impair the regenerative ability of the adipocytes (Lakowa et al., 2015; Tchkonia et al., 2013).

Cellular senescence has been hypothesized to be involved in the onset and amplification of WAT inflammageing, as in the observations and hypothesis of Tchkonia et al. (2010), which described an increase in cellular senescence in preadipocytes and adipocytes of old rats, compared to young littermates.

Cellular senescence is an irreversible cell fate, in which cells stop dividing in response to an insult such as inflammation and metabolic stress (Tchkonia et al., 2013). Senescent cells are characterized by an enlarged phenotype with a positivity for beta galactosidase, and can secrete a multitude of chemokines, cytokines, growth factors, and matrix metalloproteinases, globally defined as Senescence Associated Secretory Phenotype (SASP) (Tchkonia et al., 2013; Coppé et al., 2010).

Senescent cells are usually cleared by the immune system, however above a threshold burden, the capacity of the immune system to remove them is limited (Tchkonia et al., 2013; Wissler Gerdes et al., 2020). Thus, overexpression of senescent cells in WAT with aging amplifies its inflammation and alters the production and structure of the extracellular matrix.

Moreover, cellular senescence in WAT has been associated with impaired adipogenesis and an increase in lipotoxicity as senescent adipocytes fail to sequester lipotoxic fatty acids (Tchkonia et al., 2010). In fact in co-culture experiments, senescent cells have been shown to affect the function of adipose-derived progenitors and to alter the insulin sensitivity of WAT (Xu et al., 2015). In addition to these paracrine effects, adipose tissue senescence also triggers systemic inflammation as well as insulin resistance in other metabolic organs (Tchkonia et al., 2010).

The role of senescence in influencing WAT physiology during aging, has been demonstrated in experiments where the effects of cellular senescence were blocked or removed (Xu et al., 2015). Xu et al. (2015) showed that the clearance of senescent cells from the WAT of old mice was associated with an improvement in adipogenesis as well as in insulin sensitivity (Xu et al., 2015). Moreover the genetic clearance of senescent cells in mice was shown to reverse WAT dysfunction with aging and to contrast the loss of subcutaneous adipose tissue typical of aging (Baker et al., 2011).

In the last few years strategies for selective elimination of senescent cells have been proposed and are currently under investigation (Wissler Gerdes et al., 2020; Tchkonia and Kirkland, 2018). In particular, it has been hypothesized that this strategy could delay or contrast the onset of chronic age-related diseases (Palmer and Kirkland, 2016; Khosla et al., 2020). Some pharmacologic compounds have been developed in the past few years or investigated for their potential to clear senescent cells selectively in different tissues. These molecules are called senolytics and have been defined on the basis of their mechanism of action to induce apoptosis in aged cells selectively (Tchkonia and Kirkland, 2018; Khosla et al., 2020; Wissler Gerdes et al., 2020). Dasatinib and quercetin were found to be able to reduce senescent cell burden in multiple tissues, and improve function in naturally aged animals (Wissler Gerdes et al., 2020).

We have recently shown that treatment with quercetin in aged preadipocyte and adipocyte cultures determined a significant decrease in the number of senescent cells, along with the suppression of ROS and SASP; moreover quercetin treatment decreased miR-155-5p expression, with downregulation of NF-κB expression and upregulation of SIRT-1, in both models, confirming a senolytic and anti-inflammatory role for quercetin not only in aged adipocytes but also for preadipocytes (Zoico unpublished results 2020).

To date, senolytic activity has been demonstrated for some compounds, mostly in the oncological context and for some tissues as well as for AT (Khosla et al., 2020; Wissler Gerdes et al., 2020). However, the use of senolytic compounds has been limited to clinical trials, since more information about the safety, tolerability and side effects of these drugs are still necessary.

4. Conclusions

Sterile inflammation, or the presence of inflammation in the absence of a known identifiable infection is a common feature of aging, a condition that was defined years ago by Franceschi et al., as inflammageing (Franceschi, 2007; Franceschi et al., 2018). WAT may be a major contributor to inflammageing in the elderly (Zamboni et al., 2014; Stout et al., 2016). This may depend on the fact that WAT has recognized immunological functions (Kershaw and Kershaw and Flier, 2004; Grant and Dixit, 2015; Frasca and Blomberg, 2020), is the largest organ of our body (Cinti, 2012), and that WAT percentage increases at 70 years of age or more (Prentice and Jebb, 2001).

Increase in VAT and ectopic fat as well decline in SAT (in particular gluteofemoral SAT), seem to be the key contributors to inflammageing (Lakowa et al., 2015). SAT incompetence may be responsible for greater VAT and ectopic fat deposition. The protective role of SAT has been recognized for many years: SAT has been found to be negatively associated with metabolic syndrome in the HABC study (Goodpaster et al., 2005), and hip circumference, a well-known anthropometric index of gluteofemoral SAT, has been shown to be negatively associated with myocardial mortality (Yusuf et al., 2005). However, hip decline across aging should be carefully monitored as a marker of poor health, in terms of inflammageing and greater morbidity and mortality, in the elderly.

Since there has been a link observed between WAT dysfunction, aging, inflammageing, and age-related chronic diseases, WAT should be considered as a therapeutic target. Targeting specific cell types in WAT could be more effective than targeting the whole WAT (Liu et al., 2020). Development of agents able to preserve and/or restore preadipocyte function in later life, to remove senescent cells and to alleviate WAT inflammation could be possible treatment options. Some pharmacological approaches have been already evaluated in animals, but studies are needed in humans even in order to optimize dosage and timing.

Decline in BAT and browning white adipocytes across aging could be also taken into consideration as a possible target of intervention.

Declaration of competing interest

None.

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References

Alt, E.U., Senst, C., Murthy, S.N., Slakey, D.P., Dupin, C.L., Chaffin, A.E., Kadowitz, P.J., Izadpanah, R., 2012. Aging alters tissue resident mesenchymal stem cell properties. Stem Cell Res. 8 (2), 215–225.

Antony, A.K., Lian, Z., Wu, H., 2018. T cells in adipose tissue in aging. Front. Immunol. 9, 2945.

Arai, Y., Kamide, K., Hirose, N., 2019. Adipokines and aging: findings from centenarians and the very old. Front. Endocrinol. 10, 142.

Atzmon, G., Pollin, T.I., Crandall, J., Tanner, K., Schechter, C.B., Scherer, P.E., Rincon, M., Siegel, G., Katz, M., Lipton, R.B., Shuldiner, A.R., Barzilai, N., 2008. Adiponectin levels and genotype: a potential regulator of life span in humans. J. Gerontol. A Biol. Sci. Med. Sci. 63 (5), 447–453.

Baker, D.J., Wijshake, T., Tchkonia, T., LeBrasseur, N.K., Childs, B.G., van de Sluis, B., Kirkland, J.L., van Deursen, J.M., 2011. Clearance of p16 Ink4a -positive senescent cells delays ageing-associated disorders. Nature 479 (7372), 232–236.

Bapat, S.P., Myoung, Suh J., Fang, S., Liu, S., Zhang, Y., Cheng, A., Zhou, C., Liang, Y., LeBlanc, M., Liddle, C., Atkins, A.R., Yu, R.T., Downes, M., Evans, R.M., Zheng, Y., 2015. Depletion of fat resident T reg cells prevents age-associated insulin resistance. Nature 528 (7580), 137–141.

Bennett R., Hamel F., Simpson R.L., Agoulnik A.I., 2019. Development of adipose fibrosis and dysfunction in aged Rxfp1$^{-/-}$ mice. Diabetes 68 (Supplement 1): 1769-P.

Blogowski, W., Ratajczak, M.Z., Zyżniewska-Banaszak, E., Dołęgowska, B., Starzyńska, T., 2012. Adipose tissue as a potential source of hematopoietic stem/progenitor cells. Obesity (Silver Spring) 20 (5), 923–931.

Bloom, S.I., Tuluca, A., Ives, S.J., Reynolds, T.H., 2020. High-fat diet induced obesity and age influence the telomere shelterin complex and telomerase gene expression in mouse adipose tissue. Physiol Rep. 8 (11), e14461.

Bosello, O., Zamboni, M., 2000. Visceral obesity and metabolic syndrome. Obes. Rev. 1 (1), 47–56.

Buffa, R., Floris, G.U., Putzu, P.F., Marini, E., 2011. Body composition variations in ageing. Coll. Antropol. 35 (1), 259–265.

Camell, C., Dixit, V.D., 2019. Aging induces Nlrp3 inflammasome-dependent adipose B cell expansion to impair metabolic homeostasis. Innov. Aging 3 (1), 106.

Carter, S., Caron, A., Richard, D., Picard, F., 2013. Role of leptin resistance in the development of obesity in older patients. Clin. Interv. Aging 8, 829–844.

Caso, G., McNurlan, M.A., Mileva, I., Zemlyak, A., Mynarcik, D.C., Gelato, M.C., 2013. Peripheral fat loss and decline in adipogenesis in older humans. Metabolism 62 (3), 337–340.

Cawthorn, W.P., Scheller, E.L., MacDougald, O.A., 2012. Adipose tissue stem cells meet preadipocyte commitment: going back to the future. J. Lipid Res. 53, 227–246.

Chimin, P., Andrade, M.L., Belchior, T., Paschoal, V.A., Magdalon, J., Yamashita, A.S., Festuccia, W.T., 2017. Adipocyte mTORC1 deficiency promotes adipose tissue inflammation and NLRP3 inflammasome activation via oxidative stress and de novo ceramide synthesis. J. Lipid Res. 58 (9), 1797–1807.

Chung, K.-J., Nati, M., Chavakis, T., Chatzigeorgiou, A., 2018. Innate immune cells in the adipose tissue. Reviews in Endocrine and Metabolic Disorders 19, 283–292.

Cinti, S., 2012. The adipose organ at a glance. Dis. Model. Mech. 5, 588–594.

Cinti, S., Mitchell, G., Barbatelli, G., Murano, I., Ceresi, E., Faloia, E., Wang, S., Fortier, M., Greenberg, A.S., Obin, M.S., 2005. Adipocyte death defines macrophage localization and function in adipose tissue of obese mice and humans. J. Lipid Res. 46 (11), 2347–2355.

Coelho, M., Oliveira, T., Fernandes, R., 2013. Biochemistry of adipose tissue: an endocrine organ. Arch. Med. Sci. 9 (2), 191–200.

Coppé, J.P., Desprez, P.Y., Krtolica, A., Campisi, J., 2010. The senescence-associated secretory phenotype: the dark side of tumor suppression. Annual Review of Pathology Mechanisms of Disease 5 (1), 99–118.

Cox, A.R., Chernis, N., Masschelin, P.M., Hartig, S.M., 2019. Immune cells gate white adipose tissue expansion. Endocrinology 160 (7), 1645–1658.

De Carvalho, F.G., Justice, J.N., de Freitas, E.C., Kershaw, E.E., Sparks, L.M., 2019. Adipose tissue quality in aging: how structural and functional aspects of adipose tissue impact skeletal muscle quality. Nutrients 11, 2553.

Donato, A.J., Henson, G.D., Hart, C.R., Layec, G., Trinity, J.D., Bramwell, R.C., Ryley, A. E., Morgan, R.G., Reihl, K.D., Hazra, S., Walker, A.E., Richardson, R.S., Lesniewski, L.A., 2014. The impact of ageing on adipose structure, function and vasculature in the B6D2F1 mouse: evidence of significant multisystem dysfunction. J. Physiol. 592 (18), 4083–4096.

Engin A., 2017. Adipose tissue hypoxia in obesity and its impact on preadipocytes and macrophages: hypoxia hypothesis. In: Engin A., Engin A. (eds) Obesity and Lipotoxicity. Advances in Experimental Medicine and Biology, vol 960. Springer, Cham.

Fernando, R., Wardelmann, K., Deubel, S., Kehm, R., Jung, T., Mariotti, M., Vasilaki, A., Gladyshev, V.N., Kleinridders, A., Grune, T., Castro, J.P., 2020. Low steady-state oxidative stress inhibits adipogenesis by altering mitochondrial dynamics and decreasing cellular respiration. Redox Biol. 32, 101507.

Findeisen, H.M., Pearson, K.J., Gizard, F., Zhao, Y., Qing, H., Jones, K.L., Cohn, D., Heywood, E.B., de Cabo, R., Bruemmer, D., 2011. Oxidative stress accumulates in adipose tissue during aging and inhibits adipogenesis. PLoS One 6 (4).

Franceschi, F., 2007. Inflammageing as a major characteristic of old people: can it be prevented or cured? Nutr. Rev. 65 (12 Pt 2), S173–S176.

Franceschi, C., Campisi, J., 2014. Chronic inflammation (inflammageing) and its potential contribution to age-associated diseases. The Journals of Gerontology: Series A 69 (S1), S4–S9.

Franceschi, C., Garagnani, P., Parini, P., Giuliani, C., Santoro, A., 2018. Inflammageing: a new immune-metabolic viewpoint for age-related diseases. Nat Rev Endocrinol. 14 (10), 576–590.

Frasca, D., Blomberg, B.B., 2020. Adipose tissue: a tertiary lumphoid organ: does it change with age? Gerontology. 66, 114–121.

Gabriely, I., Ma, X.H., Yang, X.M., Rossetti, L., Barzilai, N., 2002. Leptin resistance during aging is independent of fat mass. Diabetes 51 (4), 1016–1021.

Gardner J.P., Li S., Srinivasan S.R., Chen W., Kimura M., Lu X., Berenson G.S., Aviv A., 2005. Rise in insulin resistance is associated with escalated telomere attrition. Circulation. 3; 111(17): 2171-2177.

Garg, S.K., Delaney, C., Shi, H., Yung, R., 2014. Changes in adipose tissue macrophages and T cells during aging. Crit. Rev. Immunol. 34 (1).

Ghosh, A.K., Mau, T., O'Brien, M., Garg, S., Yung, R., 2016. Impaired autophagy activity is linked to elevated ER-stress and inflammation in aging adipose tissue. Aging 8 (10), 2525–2536.

Giordano, A., Murano, I., Mondini, E., Perugini, J., Smorlesi, A., Severi, I., Barazzoni, R., Scherer, P.E., Cinti, S., 2013. Obese adipocytes show ultrastructural features of stressed cells and die of pyroptosis. J. Lipid Res. 54 (9), 2423–2436.

Goodpaster, B.H., Krishnaswami, S., Harris, T.B., Katsiaras, A., Kritchevsky, S.B., Simonsick, E.M., Nevitt, M., Holvoet, P., Newman, A.B., 2005. Obesity, regional body fat distribution, and the metabolic syndrome in older men and women. Arch. Intern. Med. 165 (7), 777–783.

Grant, R.W., Dixit, V.D., 2015. Adipose tissue as an immunological organ. Obesity 23 (3), 512–518.

Guo, W., Pirtskhalava, T., Tchkonia, T., Xie, W., Thomou, T., Han, J., Wang, T., Wong, S., Cartwright, A., Hegardt, F.G., Corkey, B.E., Kirkland, J.L., 2007. Aging results in paradoxical susceptibility of fat cell progenitors to lipotoxicity. Am. J. Physiol. Endocrinol. Metab. 292 (4), E1041–E1051.

Guzik, T.J., Skiba, D.S., Touyz, R.M., Harrison, D.G., 2017. The role of infiltrating immune cells in dysfunctional adipose tissue. Cardiovasc. Res. 113 (9), 1009–1023.

Haigis, M.C., Sinclair, D.A., 2010. Mammalian sirtuins: biological insights and disease relevance. Annu. Rev. Pathol. 5, 253–295.

Houstis, N., Rosen, E.D., Lander, E.S., 2006. Reactive oxygen species have a causal role in multiple forms of insulin resistance. Nature 440 (7086), 944–948.

Houtkooper, R.H., Pirinen, E., Auwerx, J., 2012. Sirtuins as regulators of metabolism and healthspan. Nat Rev Mol Cell Biol. 13 (4), 225–238.

Jang, J.E., Ko, M.S., Yun, J.-Y., Kim, M.-O., Kim, J.H., Park, H.S., Kim, A.R., Kim, H.J., Kim, B.J., Ahn, Y.E., Oh, J.S., Lee, W.J., Harris, R.A., Koh, E.H., Lee, K.U., 2016. Nitric oxide produced by macrophages inhibits adipocyte differentiation and promotes profibrogenic responses in preadipocytes to induce adipose tissue fibrosis. Diabetes 65 (9), 2516–2528.

Johnson, K.O., Shannon, O.M., Matu, nj., Holliday, A., Ispoglou, T., Deighton, K., 2020. Differences in circulating appetite-related hormone concentrations between younger and older adults: a systematic review and meta-analysis. Aging Clin. Exp. Res. 32 (7), 1233–1244.

Kershaw, E.E., Flier, J.S., 2004. Adipose tissue as an endocrine organ. J. Clin. Endocrinol. Metab. 89 (6), 2548–2556.

Khan S., Tsai S., Winer D.A., 2019. Adipose tissue B cells come of age: the AABs of fat inflammation. Cell Metab. 3; 30(6): 997-999.

Khosla S., Farr J.N., Tchkonia T, Kirkland J.L, 2020. The role of cellular senescence in ageing and endocrine disease. Nat Rev Endocrinol. 16 (5): 263–275.

Kirkland, J.L., Dobson, D.E., 1997. Preadipocyte function and aging: links between age-related changes in cell dynamics and altered fat tissue function. J. Am. Geriatr. Soc. 45 (8), 959–967.

Kirkland, J.L., Tchkonia, T., Pirtskhalava, T., Han, J., Karagiannides, I., 2002. Adipogenesis and aging: does aging make fat go MAD? Exp. Gerontol. 37 (6), 757–767.

Kizer, J.R., Arnold, A.M., Strotmeyer, E.S., Ives, D.G., Cushman, M., Ding, J., Kritchevsky, S.B., Chaves, P.H., Hirsch, C.H., Newman, A.B., 2010. Change in circulating adiponectin in advanced old age: determinants and impact on physical function and mortality. The Cardiovascular Health Study All Stars Study. J. Gerontol. A Biol. Sci. Med. Sci. 65 (11), 1208–1214.

Kizer, J.R., Benkeser, D., Arnold, A.M., Mukamal, K.J., Ix, J.H., Zieman, S.J., Siscovick, D.S., Tracy, R.P., Mantzoros, C.S., Defilippi, C.R., Newman, A.B., Djousse, L., 2012. Associations of total and high-molecular-weight adiponectin with all-cause and cardiovascular mortality in older persons: the Cardiovascular Health Study. Circulation. 126 (25), 2951–2961.

Lakowa, N., Trieu, N., Flehmig, G., Lohmann, T., Schön, M.R., Dietrich, A., Zeplin, P.H., Langer, S., Stumvoll, M., Blüher, M., Klöting, N., 2015. Telomere length differences between subcutaneous and visceral adipose tissue in humans. Biochem. Biophys. Res. Commun. 457 (3), 426–432.

Liberale, L., Montecucco, F., Tardif, J.-C., Libby, P., Camici, G.G., 2020. Inflamm-ageing: the role of inflammation in age-dependent cardiovascular disease. Eur. Heart J. 41 (31), 2974–2982.

Liu Z., Wu K.K.L., Jiang X., Xu A., Cheng K.K.Y., 2020. The role of adipose tissue senescence in obesity- and ageing-related metabolic disorders. Clin Sci (Lond). 31; 134(2): 315-330.
Lumeng, C.N., Liu, J., Geletka, L., Delaney, C., Delproposto, J., Desai, A., et al., 2011. Aging is associated with an increase in T cells and inflammatory macrophages in visceral adipose tissue. J. Immunol. 187 (12), 6208–6216.
Mancuso, P., Bouchard, B., 2019. The impact of aging on adipose function and adipokine synthesis. Front. Endocrinol. 10, 137.
Mazzali, G., Di Francesco, V., Zoico, E., Fantin, F., Zamboni, G., Benati, C., Bambara, V., Negri, M., Bosello, O., Zamboni, M., 2006. Interrelations between fat distribution, muscle lipid content, adipocytokines, and insulin resistance: effect of moderate weight loss in older women. Am. J. Clin. Nutr. 84 (5), 1193–1199.
Mendes, K.L., de Farias Lelis, D., Sousa Santos, S.H., 2017. Nuclear sirtuins and inflammatory signaling pathways. Cytokine Growth Factor Rev. 38, 98–105.
Minamino, T., Orimo, M., Shimizu, I., Kunieda, T., Yokoyama, M., Ito, T., Nojima, A., Nabetani, A., Oike, Y., Matsubara, H., Ishikawa, F., Komuro, I., 2009. A crucial role for adipose tissue p53 in the regulation of insulin resistance. Nat. Med. 15 (9), 1082–1087.
Moreno-Navarrete, J.M., Fernandez-Real, J.M., 2019. The gut microbiota modulates both browning of white adipose tissue and the activity of brown adipose tissue. Reviews in Endocrine and Metabolic Disorders 20, 387–397.
Ota, H., Akishita, M., Eto, M., Iijima, K., Kaneki, M., Ouchi, Y., 2007. Sirt1 modulates premature senescence-like phenotype in human endothelial cells. J. Mol. Cell. Cardiol. 43 (5), 571–579.
Palmer, A.K., Kirkland, J.L., 2016. Aging and adipose tissue: potential interventions for diabetes and regenerative medicine. Exp. Gerontol. 86, 97–105.
Pararasa, C., Bailey, C.J., Griffiths, H.R., 2015. Ageing, adipose tissue, fatty acids and inflammation. Biogerontology 16 (2), 235–248.
Pasarica, M., Sereda, O.R., Redman, L.M., Albarado, D.C., Hymel, D.T., Roan, L.E., Rood, J.C., Burk, D.H., Smith, S.R., 2009. Reduced adipose tissue oxygenation in human obesity: evidence for rarefaction, macrophage chemotaxis, and inflammation without an angiogenic response. Diabetes 58 (3), 718–725.
Picard, F., Guarente, L., 2005. Molecular links between aging and adipose tissue. Int. J. Obes. 29 (Suppl. 1), S36–S39.
Pischon, T., Girman, C.J., Hotamisligil, G.S., Rifai, N., Hu, F.B., Rimm, E.B., 2004. Plasma adiponectin levels and risk of myocardial infarction in men. JAMA 291, 1730–1737.
Pischon, T., Hu, F.B., Girman, C.J., Rifai, N., Manson, J.E., Rexrode, K.M., Rimm, E.B., 2011. Plasma total and high molecular weight adiponectin levels and risk of coronary heart disease in women. Atherosclerosis 219, 322–329.
Poehls, J., Wassel, C.L., Harris, T.B., Havel, P.J., Swarbrick, M.M., Cummings, S.R., Newman, A.B., Satterfield, S., Kanaya, A.M., 2009. Association of adiponectin with mortality in older adults: the health, aging, and body composition study. Diabetologia 52, 591–595.
Polonis, K., Becari, C., Chahal, C.A.A., Zhang, Y., Allen, A.M., Kellogg, T.A., Somers, V.K., Singh, P., 2020. Chronic intermittent hypoxia triggers a senescence-like phenotype in human white preadipocytes. Sci. Rep. 10 (1), 6846.
Prentice, A.M., Jebb, S.A., 2001. Beyond body mass index. Obes. Rev. 2 (3), 141–147.
Pyrina, I., Chung, K.-J., Michailidou, Z., Koutsilieris, M., Chavakis, T., Chatzigeorgiou, A., 2020. Fate of adipose progenitor cells in obesity-related chronic inflammation. Front. Cell Dev. Biol. 8, 644.
Rossi, A.P., Fantin, F., Zamboni, G.A., Mazzali, G., Rinaldi, C.A., Giglio, M.D., Di Francesco, V., Barillari, M., Pozzi, Mucelli R., Zamboni, M., 2011. Predictors of ectopic fat accumulation in liver and pancreas in obese men and women. Obesity 19 (9), 1747–1754.
Saely, C.H., Geiger, K., Drexel, H., 2012. Brown versus white adipose tissue: a mini-review. Gerontology 58 (1), 15–23.
Scarpace, P.J., Tümer, N., 2001. Peripheral and hypothalamic leptin resistance with age-related obesity. Physiol. Behav. 74 (4–5), 721–727.
Schipper, B., Marra, K.G., Zhang, W., Donnenberg, A.D., Rubin, J.P., 2008. Regional anatomic and age effects on cell function of human adipose-derived stem cells. Ann. Plast. Surg. 60 (5), 538–544.
Schug, T.T., Li, X., 2011. Sirtuin 1 in lipid metabolism and obesity. Ann. Med. 43 (3), 198–211.
Senesi, L., De Francesco, F., Farinelli, L., Manzotti, S., Gagliardi, G., Papalia, G.F., Riccio, M., Gigante, A., 2019. Mechanical and enzymatic procedures to isolate the stromal vascular fraction from adipose tissue: preliminary results. Front. Cell Dev. Biol. 7, 88.
Sepe, A., Tchkonia, T., Thomou, T., Zamboni, M., Kirkland, J.L., 2011. Aging and regional differences in fat cell progenitors - a mini-review. Gerontology 57 (1), 66–75.
Skurk, T., Alberti-Huber, C., Herder, C., Hauner, H., 2007. Relationship between adipocyte size and adipokine expression and secretion. J. Clin. Endocrinol. Metab. 92 (3), 1023–1033.
Stout, M.B., Justice, J.N., Nicklas, B.J., Kirkland, J.L., 2016. Physiological aging: links among adipose tissue dysfunction, diabetes, and frailty. Physiology. 32 (1), 9–19.
Suganami, T., Nishida, J., Ogawa, Y., 2005. A paracrine loop between adipocytes and macrophages aggravates inflammatory changes: role of free fatty acids and tumor necrosis factor alpha. Arterioscler. Thromb. Vasc. Biol. 25 (10), 2062–2608.
Sun, K., Tordjman, J., Clément, K., Scherer, P.E., 2013. Fibrosis and adipose tissue dysfunction. Cell Metab. 18 (4), 470–477.
Tardif, N., Salles, J., Guillet, C., Tordjman, J., Reggio, S., Landrier, J.-F., Giraudet, C., Patrac, V., Bertrand-Michel, J., Migne, C., Collin, M.L., Chardigny, J.M., Boirie, Y., Walrand, S., 2014. Muscle ectopic fat deposition contributes to anabolic resistance in obese sarcopenic old rats through eIF2α activation. Aging Cell 13 (6), 1001–1011.
Tchkonia, T., Kirkland, J.L., 2018. Aging, cell senescence, and chronic disease: emerging therapeutic strategies. JAMA 320 (13), 1319–1320.
Tchkonia, T., Pirtskhalava, T., Thomou, T., Cartwright, M.J., Wise, B., Karagiannides, I., Shpilman, A., Lash, T.L., Becherer, J.D., Kirkland, J.L., 2007. Increased TNFα and CCAAT/enhancer-binding protein homologous protein with aging predispose preadipocytes to resist adipogenesis. Am J Physiol-Endocrinol Metab. 293 (6), E1810–E1819.
Tchkonia, T., Morbeck, D.E., Zglinicki, T.V., Deursen, J.V., Lustgarten, J., Scrable, H., Khosla, S., Jensen, M.D., Kirkland, J.L., 2010. Fat tissue, aging, and cellular senescence. Aging Cell 9 (5), 667–684.
Tchkonia, T., Zhu, Y., van Deursen, J., Campisi, J., Kirkland, J.L., 2013. Cellular senescence and the senescent secretory phenotype: therapeutic opportunities. J. Clin. Invest. 123 (3), 966–972.
Thomas, E.L., Parkinson, J.R., Frost, G.S., Goldstone, A.P., Doré, C.J., McCarthy, J.P., Collins, A.L., Fitzpatrick, J.A., Durighel, G., Taylor-Robinson, S.D., Bell, J.D., 2012. The missing risk: MRI and MRS phenotyping of abdominal adiposity and ectopic fat. Obesity 20, 76–87.
Trim, W., Turner, J.E., Thompson, D., 2018. Parallels in immunometabolic adipose tissue dysfunction with ageing and obesity. Front. Immunol. 9, 169.
Tzanetakou, I.P., Katsilambros, N.L., Benetos, A., Mikhailidis, D.P., Perrea, D.N., 2012. "Is obesity linked to aging?": adipose tissue and the role of telomeres. Ageing Res. Rev. 11 (2), 220–229.
Unger, R.H., 2005. Longevity, lipotoxicity and leptin: the adipocyte defense against feasting and famine. Biochimie 87, 57–64.
Vergoni, B., Cornejo, P.J., Gilleron, J., Djedaini, M., Ceppo, F., Jacquel, A., Bouget, G., Ginet, C., Gonzalez, T., Maillet, J., Dhennin, V., Verbanck, M., Auberger, P., Froguel, P., Tanti, J.F., Cormont, M., 2016. DNA damage and the activation of the p53 pathway mediate alterations in metabolic and secretory functions of adipocytes. Diabetes. 65 (10), 3062–3074.
Wang, H., Shen, L., Sun, X., Liu, F., Feng, W., Jiang, C., Chu, X., Ye, X., Jiang, C., Wang, Y., Zhang, P., Zang, M., Zhu, D., Bi, Y., 2019. Adipose group 1 innate lymphoid cells promote adipose tissue fibrosis and diabetes in obesity. Nat. Commun. 10 (1), 3254.
Wannamethee, S.G., Whincup, P.H., Lennon, L., Sattar, N., 2007. Circulating adiponectin levels and mortality in elderly men with and without cardiovascular disease and heart failure. Arch. Intern. Med. 167, 1510–1517.
Wissler Gerdes, E.O., Zhu, Y., Tchkonia, T., Kirkland, J.L., 2020. Discovery, development, and future application of senolytics: theories and predictions. FEBS J. 287 (12), 2418–2427.
Xu, M., Palmer, A.K., Ding, H., Weivoda, M.M., Pirtskhalava, T., White, T.A., Sepe, A., Johnson, K.O., Stout, M.B., Giorgadze, N., Jensen, M.D., LeBrasseur, N., Tchkonia, T., Kirkland, J.L., et al., 2015. Targeting senescent cells enhances adipogenesis and metabolic function in old age. eLife 4, e12997.
Yang, W., Yang, C., Luo, J., Wei, Y., Wang, W., Zhong, Y., 2018. Adiponectin promotes preadipocyte differentiation via the PPARγ pathway. Mol. Med. Rep. 17 (1), 428–435.
Yu, Y.-H., Zhu, H., 2004. Chronological changes in metabolism and functions of cultured adipocytes: a hypothesis for cell aging in mature adipocytes. Am J Physiol-Endocrinol Metab. 286 (3), E402–E410.
Yusuf, S., Hawken, S., Ounpuu, S., Bautista, L., Franzosi, M.G., Commerford, P., Lang, C. C., Rumboldt, Z., Onen, C.L., Lisheng, L., Tanomsup, S., Wangai, P.J., Razak, F., M Sharma, A., Anand, S.S., INTERHEART Study Investigators, 2005. Obesity and the risk of myocardial infarction in 27,000 participants from 52 countries: a case-control study. Lancet 366 (9497), 1640–1649.
Zamboni, M., Mazzali, G., Zoico, E., Harris, T.B., Meigs, J.B., Di Francesco, V., Fantin, F., Bissoli, L., Bosello, O., 2005. Health consequences of obesity in the elderly: a review of four unresolved questions. Int. J. Obes. 29 (9), 1011–1029.
Zamboni, M., Rossi, A.P., Fantin, F., Zamboni, G., Chirumbolo, S., Zoico, E., Mazzali, G., 2014. Adipose tissue, diet and aging. Mech. Ageing Dev. 136–137, 129–137.
Zhang, L., Ebenezer, P.J., Dasuri, K., Fernandez-Kim, S.O., Francis, J., Mariappan, N., Gao, Z., Ye, J., Bruce-Keller, A.J., Keller, J.N., 2011. Aging is associated with hypoxia and oxidative stress in adipose tissue: implications for adipose function. Am J Physiol-Endocrinol Metab. 301 (4), E599–E607.
Zhang, M., Sheng, S., Zhang, W., Zhang, J., Zhang, Z., Zhang, M., Hatch, G.M., Chen, L., 2018. MiR27a promotes the development of macrophage-like characteristics in 3T3-L1 preadipocytes. Int. J. Biol. Sci. 14 (11), 1599–1609.
Zhu, Y., Liu, X., Ding, X., Wang, F., Geng, X., 2019. Telomere and its role in the aging pathways: telomere shortening, cell senescence and mitochondria dysfunction. Biogerontology. 20 (1), 1–16.
Zoico, E., Di Francesco, V., Bissoli, L., Mazzali, G., Fontana, G., Giuliano, K., Bosello, O., Zamboni, M., 2008. Interrelationships between leptin resistance, body composition, and aging in elderly women. J. Am. Geriatr. Soc. 56 (9), 1768–1769.
Zoico, E., Garbin, U., Olioso, D., Mazzali, G., Fratta Pasini, A.M., Di Francesco, V., Sepe, A., Cominacini, L., Zamboni, M., 2009. The effects of adiponectin on interleukin-6 and MCP-1 secretion in lipopolysaccharide-treated 3T3-L1 adipocytes: role of the NF-kappaB pathway. Int. J. Mol. Med. 24 (6), 847–851.
Zoico, E., Di Francesco, V., Olioso, D., Fratta Pasini, A.M., Sepe, A., Bosello, O., Cinti, S., Cominacini, L., Zamboni, M., 2010a. In vitro aging of 3T3-L1 mouse adipocytes leads to altered metabolism and response to inflammation. Biogerontology. 11 (1), 111–122.
Zoico, E., Rossi, A., Di Francesco, V., Sepe, A., Olioso, D., Pizzini, F., Fantin, F., Bosello, O., Cominacini, L., Harris, T.B., Zamboni, M., 2010b. Adipose tissue infiltration in skeletal muscle of healthy elderly men: relationships with body composition, insulin resistance, and inflammation at the systemic and tissue level. J. Gerontol. A Biol. Sci. Med. Sci. 65 (3), 295–299.

Zoico, E., Rizzatti, V., Policastro, G., Tebon, M., Darra, E., Rossi, A.P., Mazzali, G., Fantin, F., Zamboni, M., 2019a. In vitro model of chronological aging of adipocytes: interrelationships with hypoxia and oxidation. Exp. Gerontol. 121, 81–90.
Zoico, E., Rubele, S., De Caro, A., Nori, N., Mazzali, G., Fantin, F., et al., 2019b. Brown and beige adipose tissue and aging. Front. Endocrinol. 10.
Zoico, E., Policastro, G., Rizzatti, V., Nori, N., Darra, E., Rossi, A.P., Fantin, F., Zamboni, M., 2020. Mechanisms of adipose tissue extracellular matrix alterations in an *in vitro* model of adipocytes hypoxia and aging. Mechanism of Ageing and Development. https://doi.org/10.1016/j.mad.2020.111374 (in press).