SPARC: a key player in the pathologies associated with obesity and diabetes

Katarina Kos and John P. H. Wilding

Abstract | SPARC (secreted protein acidic and rich in cysteine, also known as osteonectin or BM-40) is a widely expressed profibrotic protein with pleiotropic roles, which have been studied in a variety of conditions. Notably, SPARC is linked to human obesity; SPARC derived from adipose tissue is associated with insulin resistance and secretion of SPARC by adipose tissue is increased by insulin and the adipokine leptin. Furthermore, SPARC is associated with diabetes complications such as diabetic retinopathy and nephropathy, conditions that are ameliorated in the *Sparc*-knockout mouse model. As a regulator of the extracellular matrix, SPARC also contributes to adipose-tissue fibrosis. Evidence suggests that adipose tissue becomes increasingly fibrotic in obesity. Fibrosis of subcutaneous adipose tissue may restrict accumulation of triglycerides in this type of tissue. These triglycerides are, therefore, diverted and deposited as ectopic lipids in other tissues such as the liver or as intramyocellular lipids in skeletal muscle, which predisposes to insulin resistance. Hence, SPARC may represent a novel and important link between obesity and diabetes mellitus. This Review is focused on whether SPARC could be a key player in the pathology of obesity and its related metabolic complications.

Kos, K. & Wilding, J. P. H. *Nat. Rev. Endocrinol.* **6**, 225–235 (2010); published online 2 March 2010; doi:10.1038/nrendo.2010.18

Introduction

SPARC (secreted protein acidic and rich in cysteine, also known as BM-40) was initially identified in bone, in 1981, and is thus also known as osteonectin.[1] SPARC is also highly expressed in several tumors, such as ovarian and colorectal tumors and melanomas.[2] Since these early studies SPARC was found to be ubiquitously expressed and was studied in various pathological conditions ranging from liver and kidney disease to Alzheimer disease. SPARC was also found to be secreted by adipose cells and circulating SPARC levels positively correlate with BMI in humans.[3,4] These findings suggest that the secretion of SPARC from adipose tissue may account for the majority of circulating SPARC.[3] Given the profibrotic qualities of SPARC and the evidence of fibrosis in adipose tissue of individuals with obesity, a finding reported in 2008,[5] SPARC was proposed to contribute to the pathogenesis of obesity-associated disorders, in particular by promoting insulin resistance.[3] In this Review we briefly explain the various functions of SPARC, with a focus on adipose tissue. We also examine the role of SPARC in association with type 2 diabetes mellitus (T2DM) and other obesity-related complications ranging from cardiovascular disease to cancer, and explore the potential of SPARC as a therapeutic target.

Structure and general function of SPARC

SPARC is a glycoprotein of the extracellular matrix (ECM) that binds calcium, collagen and hydroxyapatite. SPARC weighs 34 kDa, but owing to glycosylation the secreted form migrates to 43 kDa on sodium dodecyl sulfate polyacrylamide gel electrophoresis.[6] As a secreted protein, SPARC is an extracellular regulatory macromolecule that does not contribute to the matrix structure, but regulates the cell–matrix interaction.

Structure

SPARC is the product of a single gene localized on chromosomal region 5q31-33, in the proximity of the genes encoding macrophage colony-stimulating factor 1 (CSF1), interleukin 3 (IL-3), platelet-derived growth factor (PDGF) and the β2 adrenergic receptor.[7] The sequences of the *SPARC* gene and the protein it encodes are highly conserved among species.[8] The structure of the human protein consists of 286 residues and three domains (Figure 1). The N-terminal domain of SPARC is an acidic region that contains the major immunological epitopes of the protein.[9] The second domain is a cysteine-rich follistatin-like domain, which binds activin, inhibin, heparin and proteoglycans and may regulate proliferation of endothelial cells.[8] The C-terminal domain is a calcium-binding extracellular domain, which includes a binding site that interacts with endothelial cells and binds fibril-forming collagens (the latter occurring with increased affinity after cleavage of a single-bond peptide by metalloproteinases).[10–12]

General function

SPARC is counteradhesive (disrupts cell adhesion), a modulator of cell-surface interaction and an inhibitor

Competing interests
The authors declare no competing interests.

Department of Diabetes and Vascular Medicine, Peninsula College of Medicine and Dentistry, University of Exeter, Peninsula Medical School Building, Barrack Road, Exeter EX2 5DW, UK (**K. Kos**). Diabetes and Endocrinology Clinical Research Unit, Clinical Sciences Center, University Hospital Aintree, Longmoor Lane, Liverpool L9 7AL, UK (**J. P. H. Wilding**).

Correspondence to:
K. Kos
katarina.kos@pcmd.ac.uk

Key points

- An increase in the levels of SPARC is found in animals and human individuals with obesity and insulin resistance
- Raised SPARC concentrations are associated with the development of diabetes-associated complications
- SPARC is involved in strengthening bone but raised concentrations of the protein are associated with increased cardiovascular risk
- Associations between SPARC and cancer have been postulated, but the exact role of this protein in tumorigenesis is unclear
- SPARC antagonism may help in the prevention of obesity-related complications

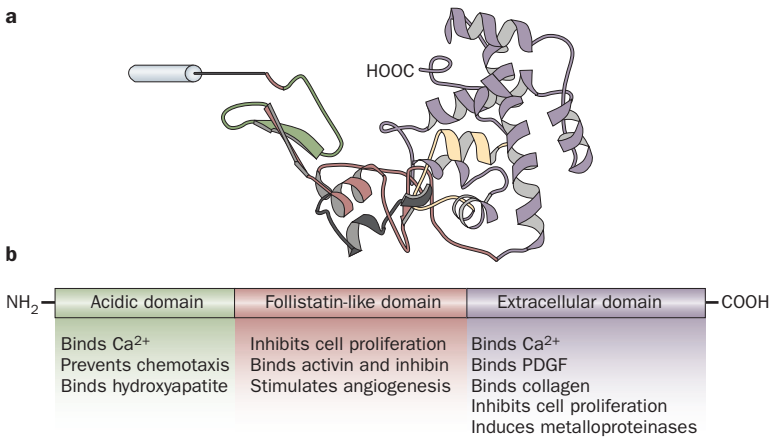

Figure 1 | Structure and functional properties of SPARC. **a** | Crystallographic structure of human SPARC. The acidic domain is shown in green, the follistatin-like domain in red and the extracellular domain in purple. **b** | Functional properties of the three protein domains. Abbreviation: SPARC, secreted protein acidic and rich in cysteine (also known as osteonectin or BM-40). Permission for part a obtained from The American Society for Clinical Investigation © Bradshaw, A. D. & Sage, E. H. *J. Clin. Invest.* **107**, 1049–1054 (2001).[111]

of the cell cycle. This protein is expressed when tissues undergo events that require changes in cell–matrix and cell–cell contact, particularly tissue renewal, tissue remodeling, and embryonic development.[13] SPARC modulates tissue physiology by altering the cell–ECM interaction, cell proliferation and cell migration. Owing to these properties, SPARC participates in wound healing, angiogenesis, tumorigenesis and inflammation.[14] These functions are facilitated by SPARC's ability to influence the activity of cytokines and growth factors, such as vascular endothelial growth factor (VEGF), which is the most potent and widely distributed angiogenic mitogen for capillary endothelial cells,[15–16] PDGF, heparin-binding growth factor 2 (HBGF2, also known as FGF2) and insulin-like growth factor 1 (IGF1).[17] Box 1 provides more details about proteins that are known to influence or are influenced by SPARC.

The regulation of *SPARC* gene expression and SPARC protein function have been studied in most detail in bone, where SPARC links organic and mineral phases of the bone ECM.[18] SPARC has also been detected in myoblasts, myotubes and muscle fibers, where its levels increase during muscle development and regeneration.[19] Adipose tissue cells are the major source of circulating SPARC in obesity and this protein has important, although not yet fully understood roles in adipose tissue physiology.[3,20]

SPARC has been detected intracellularly and extracellularly.[14] Once secreted, this protein is rapidly broken down in the circulation by extracellular proteases and its breakdown products are thought to be functional, although evidence is still lacking.[21] A putative receptor for extracellular SPARC is integrin α5β1, which activates the Wnt/β-catenin pathway.[22] The complex structure of SPARC, which contains three domains with different properties, and the fact that protein binding depends on physiological cell status and differs depending on type of tissue add to the complexity of understanding the mechanisms of action of this protein. Some studies have described post-translational tissue-specific modifications caused by disulfide crosslinks between SPARC and other cysteine-rich proteins or glycosylation of SPARC at the follistatin-like domain, which may give SPARC tissue-specific collagen-binding properties.[23] In addition, several SPARC-like proteins with functional similarities to SPARC have been identified. Of these, SPARC-like protein 1 (also known as hevin) is expressed in the human central nervous system, along with SPARC,[24] but the extent to which they share functionality is not clear. The characteristics of adipocyte-derived SPARC, including its post-translational modifications and the binding sites that are relevant for adipose tissue physiology, have not been investigated in depth. Several reviews have been published on SPARC's complex nature.[8,14,24]

Sparc-knockout mouse model

No mutations of *SPARC* have been identified in human populations. Two different mouse gene knockout strains, referred to as exon 4 or exon 6 *Sparc*-knockout lines, have been generated; in both, the SPARC protein is completely absent.[25] *Sparc*-knockout mice are characterized by low-turnover osteopenia[25] and early cataract development.[26,27] Of note, the diameter of collagen fibrils in the dermis increases and its tensile strength is reduced in these mice; collagen content is also altered, with an increase in type VI collagen and a decrease in type I collagen.[28] Furthermore, the size of subcutaneous adipose tissue pads is increased in *Sparc*-knockout mice compared with that in controls, despite similar body weight.[29] A potential explanation for this lack of difference in weight is that the bone of knockout mice is osteopenic and might be lighter than that of wild-type mice.[26]

SPARC and obesity

Obesity, defined as a BMI of more than 30 kg/m^2, is closely linked with an elevated risk of metabolic complications, particularly T2DM[30] and the metabolic syndrome (which often precedes diabetes mellitus).[31] Furthermore, individuals with obesity have an increased risk of developing cardiovascular disease,[32] which is partly due to the metabolic consequences of obesity, such as T2DM. Obesity is also linked to an increased risk of cancer and mortality caused by cancer, and is associated with various cancers, such as those in the gastrointestinal tract and endometrium.[33] SPARC dysregulation has been associated with a wide range of obesity-related disorders, including T2DM and its complications, renal and liver

disease, cardiovascular disease and obesity-associated cancers (Table 1).

The following sections describe the changes in adipose tissue associated with obesity and highlight SPARC as a potential key factor contributing to fibrosis of adipose tissue and obesity-associated complications.

Adipose tissue fibrosis

ECM changes in obesity

The composition and characteristics of the ECM in adipose tissue change with obesity. Immunostaining with the fibrosis marker picrosirius red shows more fibrosis in the subcutaneous adipose tissue of individuals with severe obesity than in that of lean controls.[5] Analysis of the transcriptomic signature of subcutaneous white adipose tissue in these individuals revealed that the adipose tissue of those with obesity had increased expression of genes coding for ECM proteins that characterize fibrosis, such as integrins (for example, the fibronectin receptor) and members of the collagen family, including the α chain of type IV collagen that is most abundant in basal membranes and fibril-associated collagens. These genes were co-expressed with genes coding for ECM modulators, such as lumican and laminin β1 chain, which have also a role in the initiation of inflammatory phenomena.[5] A study *in vitro* has shown that disturbance of the three-dimensional structure of ECM by increased fibril-forming collagen content leads to increased rigidity and compromises adipocyte differentiation.[34]

Consequences of fibrosis

An imbalance in collagen expression has been associated with metabolic dysregulation and insulin resistance in mice: for example, knockout of the *Col6a3* gene, which encodes the collagen α3(VI) chain, resulted in weakening of the extracellular scaffold, enabling an expansion of adipocytes within subcutaneous adipose tissue, accompanied by reduced adipose tissue inflammation, and protected mice from insulin resistance induced by a lipid-rich diet.[35] These findings suggest that fibrosis of the subcutaneous adipose tissue may reduce its ability to store triglycerides, which then overspill into the circulation, which results in systemic hyperlipidemia and, ultimately, lipid infiltration of other organs. Ectopic adiposity contributes to the pathophysiology of insulin resistance. This link is most apparent in patients with familial lipodystrophy, in whom storage of triglycerides in subcutaneous adipose tissue is severely compromised.[36–38]

Classical sites of ectopic lipid deposition are the skeletal muscle, in which lipids are stored within myocytes, and the liver, in which increased adiposity may lead to non-alcoholic steatohepatitis, which can progress to cirrhosis. In addition, lipids can accumulate in pancreatic β cells and impair pancreatic function. Increased lipid accumulation is also found in the myocardium, epicardium and as perivascular adipose tissue. Perivascular adipose tissue surrounds all noncentral arteries and can influence blood pressure and enhance atherosclerosis.[39,40]

In contrast to subcutaneous adipose tissue fibrosis, which limits the expansion of this tissue and leads to metabolic complications, a combination of free expansion of subcutaneous adipose tissue and reduction of ectopic lipid stores is associated with improved insulin sensitivity. This link has been experimentally demonstrated by overexpressing adiponectin in leptin-deficient *ob/ob* mice, a model of T2DM, which led to an increase in subcutaneous adipose tissue stores and, despite increased weight, improved insulin sensitivity.[41] Furthermore, individuals with large subcutaneous abdominal adipose

Box 1 | Modulators of SPARC and SPARC regulation

SPARC

Stimulators
- Insulin
- Leptin
- Heat shock
- Retinoic acid
- Dexamethasone
- Transforming growth factor β1
- Activin
- Bone morphogenetic protein 1
- Platelet-derived growth factor
- Insulin-like growth factor 1
- β-Adrenergic stimulation

Inhibitors
- Glucose
- Heparin-binding growth factor 2
- Interleukin 1
- TNF
- Phorbol-12-myristate-13-acetate
- Epidermal growth factor
- Pb^{2+}
- Platelet-derived growth factor
- Lipopolysaccharide
- Amyloid β A4 protein
- Stabilin 1

Molecules influenced by SPARC

Stimulated
- Plasminogen activator inhibitor 1
- Metalloproteinases
- Fibronectin
- Collagens
- RACα serine/threonine protein kinase (also known as C-AKT or PKB)

Inhibited
- Vascular endothelial growth factor
- Leptin
- Lipopolysaccharide
- CAAT/enhancer-binding protein α and CAAT/enhancer-binding protein β
- Peroxisome proliferator-activated receptor γ
- Laminin
- Transforming growth factor β1

Table 1 | SPARC and obesity-associated disorders

Function	Obesity-associated complication
Inhibition of adipogenesis	Ectopic lipid deposition leading to fatty liver disease, cardiovascular disease and insulin resistance
Regulation of collagen composition with profibrotic characteristics	Heart failure, premature aging, renal and hepatic fibrosis, and adipose tissue fibrosis, which leads to ectopic lipid deposition
Osteoblastogenesis	Strengthening bone composition
Reduced angiogenesis	Diabetic retinopathy and adipose tissue hypoxia
Tumorigenesis	Potentially furthers progression of obesity-associated cancers
Neuronal cell detachment and cell death	Alzheimer disease

tissue deposits have less ectopic lipid deposition than individuals with obesity who have smaller subcutaneous adipose tissue deposits, as most clearly demonstrated in patients with lipodystrophy.[42] This finding supports the concept that subcutaneous adipose tissue acts as a 'metabolic sink'.[43,44]

Inflammation

Obesity is characterized by a proinflammatory state, which may contribute to obesity-related complications. The circulating levels of markers of systemic inflammation are increased in obesity and this effect has been attributed to the increased adipose tissue mass. These markers include adipokines (proteins secreted by adipocytes that have paracrine and endocrine function), such as leptin, and cytokines, such as tumor necrosis factor (TNF), C-reactive protein (CRP), plasminogen activator inhibitor 1 (PAI1) and interleukins.[45] The latter are secreted by stromal cells such as macrophages, which infiltrate the adipose tissue in obesity, and by adipocytes.[46]

Different adipose tissue compartments and/or lipid depots differ in endocrine activity and contribution to a proinflammatory state. The amount of visceral (omental) adipose tissue, which is the inner abdominal adipose compartment, has a stronger association with obesity-related metabolic dysfunction, such as insulin resistance and cardiovascular risk, than subcutaneous adipose tissue, but ectopic adiposity (lipid infiltration of various organs) may play an even more important role in this pathologic state.[47,48] As visceral adipose tissue expansion is enhanced when the subcutaneous adipose tissue becomes unable to store further triglycerides, we argue that it could in fact be considered as a site of ectopic lipid deposition.

Although the direct initiating causes of obesity-induced changes in the ECM that progress to adipose tissue fibrosis are unclear, studies of obesity-associated genes have suggested that a proinflammatory environment may lead to excessive synthesis of ECM components.[5,49] The secretion of chemoattractants, such as macrophage migration inhibitory factor (MIF), by adipocytes controls infiltration of the adipose tissue by macrophages, and infiltrating macrophages contribute to a proinflammatory state in individuals with obesity.[49,50]

Hypoxia

Apart from inflammatory signals, hypoxia (reduced tissue oxygenation) has also been implicated in the induction of adipose tissue fibrosis.[51] Hypoxia of adipose tissue has been observed *in vivo* in obese rodents[52,53] and also in humans with obesity.[54] Reduced partial pressure of oxygen in subcutaneous abdominal adipose tissue was associated with changes in collagen expression and decreased VEGF expression, which suggests that capillary dropout is taking place. Capillary rarefaction in the adipose tissue may, therefore, contribute to hypoxia, as this type of tissue expands in obesity.[54]

Hypoxia activates the expression of hypoxia-inducible factor 1α (HIF1α) and inflammatory cytokines in adipose tissue[55] and induces macrophage infiltration, all of which increase inflammation, and can, therefore, lead to fibrosis.[49,55] The expression of *HIF1A* (the gene that encodes HIF1α) in adipose tissue increases with weight gain and decreases with weight loss.[51,56]

In addition to adipocytes, adipose tissue consists of adipose stromal cells that include adipocyte precursors, which are mesenchymal preadipocytes derived from stem cells, and also macrophages, endothelial cells and a few smooth muscle cells. Macrophages induce inflammation, but also release products that prime human preadipocytes to acquire a profibrotic phenotype and stimulate the synthesis of several ECM proteins from these profibrotic cells.[49] Preadipocytes secret SPARC during their differentiation, but little is known about the contribution of stromal cells to the maintenance of the ECM during adipose tissue fibrosis.

Role of SPARC in adipose tissue

SPARC is secreted by most tissues but, in obesity, the protein is predominantly secreted by adipose tissue.[3,20] SPARC is detected at much higher levels in subcutaneous abdominal adipose tissue than in visceral adipose tissue and is mainly produced by adipocytes, rather than by stromal vascular cells.[3,57] SPARC expression and secretion during adipocyte differentiation is biphasic, with the highest levels occurring in preadipocytes during the early stages of differentiation; after a decline, protein levels are again elevated in fully differentiated adipocytes.[57] SPARC secretion is increased in obese mice, including *ob/ob* mice.[20] In humans, *SPARC* mRNA expression is higher in individuals with obesity than in individuals who are not obese, increases with weight gain and decreases with dietary-induced weight loss.[3]

Inhibition of adipogenesis

SPARC limits adipogenesis by inhibiting the differentiation of preadipocytes to mature adipocytes.[58] A possible mechanism underlying inhibition of adipogenesis is the stimulation of the Wnt/β-catenin signaling pathway, which leads to an increase of osteogenesis with decreased adipogenesis. This latter effect is caused by inhibition of expression of adipogenic transcription factors, including CAAT/enhancer-binding protein α (C/EBPα), CAAT/enhancer-binding protein β (C/EBPβ) and peroxisome proliferator-activated receptor γ (PPARγ).[58] Furthermore,

independently of changes in expression of transcription factors, SPARC changes the ECM into which adipocytes are embedded by enhancing the deposition of fibronectin, but inhibiting the deposition of laminin.[58] Conversion of a fibronectin-rich ECM, where adipocyte differentiation is limited,[59] to a laminin-rich ECM enables fusiform pre-adipocytes to form spherical adipocytes.[58] The inhibition of laminin function impairs basal lamina formation and, consequently, adipogenesis.[58] SPARC causes changes on fibril formation and contractility through integrin-linked kinase activation.[60]

The lack of weight gain with increased adiposity observed in the *Sparc*-knockout mouse model may be explained by the potential for unrestrained subcutaneous adipose tissue expansion. *Sparc*-knockout mice have adipocytes of greater diameter, a larger amount of subcutaneous and epididymal adipose tissue and a greated number of adipocytes in epididymal adipose pads than wild-type mice.[29] In humans, the secretion of SPARC is higher in subcutaneous tissue, where it promotes fibrosis and inhibits adipogenesis, than in visceral adipose tissue.[3] These observations and the results from studies on *Sparc*-knockout mice suggest that SPARC may limit the expansion of subcutaneous adipose tissue also in humans.

SPARC and leptin

SPARC is linked with leptin, one of the key secretory products of adipose tissue.[3] Circulating levels of leptin are strongly associated with adipose-tissue mass, in whose regulation leptin is involved by a central feedback mechanism (Figure 2a). Elevated concentrations of leptin in the circulation and cerebrospinal fluid are sensed by the arcuate nucleus of the hypothalamus and lead to suppression of appetite; they also lead to activation of the sympathetic axis via the tractus solitarius, which induces lipolysis, and thus reduces adipose mass, in white adipose tissue.[61] Leptin secretion in human white adipose tissue is in turn downregulated by β-adrenergic activation.[62]

SPARC and leptin gene expression and protein levels are higher in subcutaneous adipose tissue than in visceral adipose tissue.[3] Human *SPARC* gene expression is upregulated by leptin *in vitro*, which may therefore contribute to elevated SPARC levels in individuals with obesity, while SPARC, in turn, reduces leptin expression (Figure 2a).[3,58] Thus, SPARC and leptin regulate each other by a local feedback loop within adipose tissue and act to counteract an increase in adipose tissue mass (Figure 2a).

SPARC levels in the heart can be experimentally induced through β-adrenergic activation,[63] and sympathetic innervation might increase SPARC secretion in adipose tissue, although this effect has not been proven. *SPARC* gene and protein expression have also been detected in the brain in mice, rats and humans,[3,64,65] but the physiological role of SPARC in the central nervous system is currently not known. In particular, the possibility that SPARC has a direct role in the regulation of energy balance has not been investigated, although its peripheral interaction with leptin supports an indirect role. SPARC is probably present in the cerebrospinal fluid, as it is secreted, among other cell types, by astrocytes and immune cells, which exist there at low concentrations. SPARC gene and protein expression were detected in cell components of the blood–brain barrier, such as ependymal cells, cells of the choroid plexus and pia mater in mice.[65] We are not aware of any work that has directly measured SPARC concentrations in the

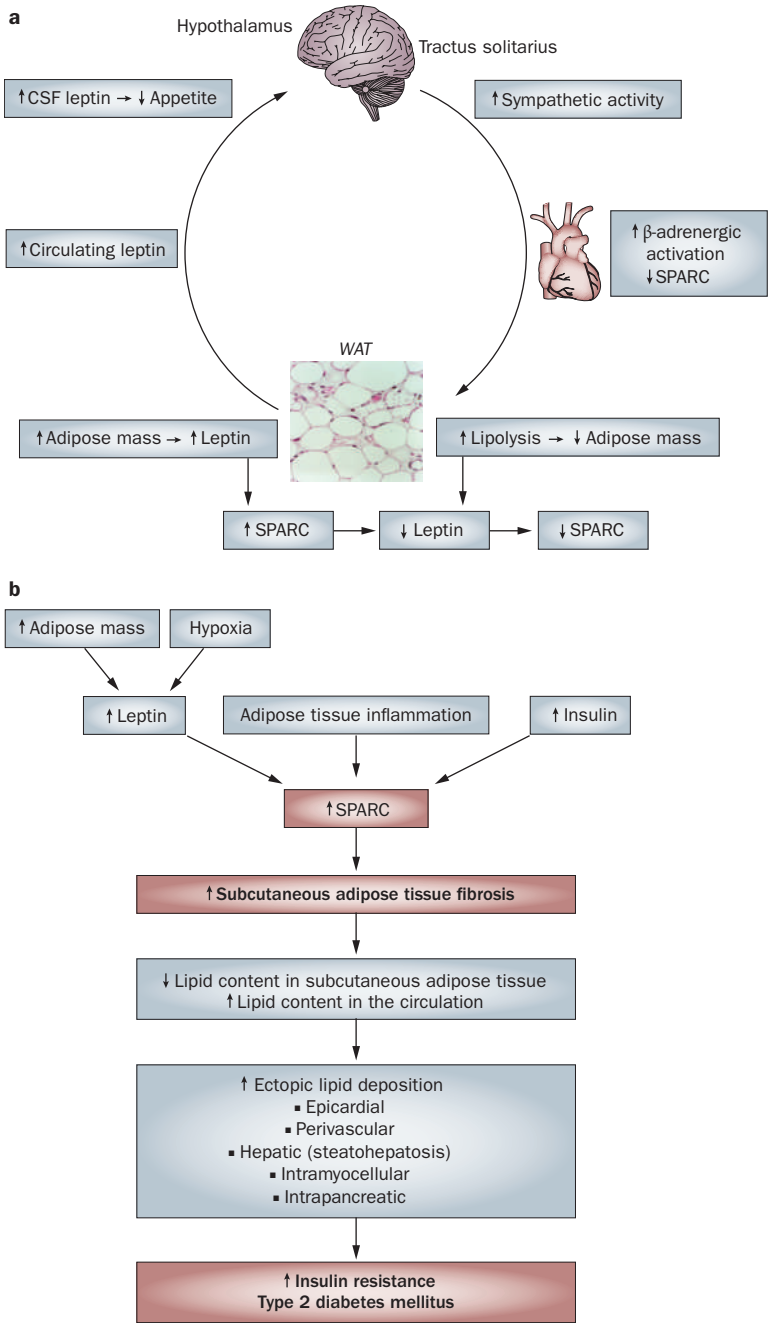

Figure 2 | Regulation of adipose mass by SPARC and leptin. **a** | SPARC and leptin regulate increases in adipose mass. Leptin and β-adrenergic stimulation are part of a central feedback loop mechanism. **b** | The increased secretion of SPARC furthers adipose tissue fibrosis, which promotes lipid deposition in various organs and contributes to insulin resistance. Lipid deposition in the pancreas also compromises the secretion of insulin. Lipid deposition and fibrosis within the pancreas are associated with type 2 diabetes mellitus. Abbreviations: CSF, cerebrospinal fluid; SPARC, secreted protein acidic and rich in cysteine (also known as osteonectin or BM-40); WAT, white adipose tissue.

cerebrospinal fluid, however, and whether SPARC can cross the blood–brain barrier is unknown.

An inverse correlation of adiponectin and SPARC levels was found in human subcutaneous adipose tissue, whereas no association was found with resistin in subcutaneous or visceral adipose tissue.[3] An interaction of SPARC with other adipokines has not been demonstrated.

Regulation of fibrosis

SPARC may be one of the key regulators of obesity-induced adipose tissue fibrosis and associated metabolic dysfunction by regulating ECM composition and adipogenesis.[58] Studies *in vitro* have shown that SPARC increases fibronectin expression in mouse white adipose tissue cells,[58] disrupts attachment of mouse and human stromal vascular adipose tissue cells to the ECM[22] and enhances fibroblast migration by stimulating fibronectin expression in mouse heart cells.[66]

SPARC expression may contribute to adipose tissue fibrosis by inducing inflammation. SPARC is positively correlated with CRP (measured by the high-sensitivity CRP test) and expression of the chemoattractant MIF in the adipose tissue. Elevated levels of SPARC are not, however, correlated with circulating TNF or IL-6 levels[3] and TNF was shown to inhibit secretion of SPARC *in vitro*.[67] Functional studies are necessary to clarify the exact role of SPARC in inflammation and its underlying regulatory pathways. Of interest, alternatively activated macrophages—which are anti-inflammatory macrophages found in lean adipose tissue—express a receptor called stabilin 1 that was shown to induce clearance and degradation of SPARC.[68] These findings suggest that reduced clearance of SPARC in obesity-induced inflammation may promote adipose tissue fibrosis.[69]

Energy restriction leads to reduction of SPARC concentrations in mice and humans and leads to a decrease of HIF1α concentrations; conversely, HIF1α levels increase with weight gain, as SPARC levels do.[3,56] A study in lung cancer cells showed an association of SPARC with EPAS1 (also known as hypoxia-inducible factor 2α, or HIF2α), which indicates a potential upregulation of SPARC by hypoxia.[70] This upregulation could be mostly indirect, by an increase in the concentration of leptin, whose expression is induced by hypoxia *in vitro* (Figure 2b).[71]

SPARC also inhibits VEGF-stimulated proliferation of endothelial cells[15] and was associated with reduced angiogenesis—defined as formation of new blood vessels from pre-existing vessels—in some cancers.[72] Some cleavage products of SPARC, however, have been shown to stimulate angiogenesis.[73] Therefore, the exact effect of SPARC on adipose microvasculature and circulation and the subsequent risk of development of hypoxia remain to be clarified.[73]

Of note, transforming growth factor β1 (TGF-β1), which is one of the major profibrotic mediators, is released predominantly by nonadipose cells in adipose tissue and is upregulated in obesity.[74] TGF-β1 increases *SPARC* expression[67] and, as SPARC, is upregulated by hypoxia, leptin and inflammatory cytokines, as shown in pulmonary and renal fibrosis.[75–77]

SPARC and obesity-related diseases

SPARC and diabetes

Less is known about changes of ECM composition in T2DM than in obesity. The subcutaneous adipose tissue of individuals with obesity is more hypoxic and has higher expression of collagen and fibronectin than that of lean individuals.[54] The hypothesis that hypoxia induces insulin resistance has been suggested,[78,79] but the adipose tissue of patients with T2DM does not seem to be more hypoxic than adipose tissue of healthy individuals,[54] though evidence from large studies is lacking. Knockout of *Col6a3* (the gene that encodes the α3 chain of type VI collagen) in mice was associated with improved insulin resistance, which suggests that collagens and ECM composition may have a role in the pathogenesis of T2DM.[35] A functional role of SPARC in this process remains to be determined. However, an association of SPARC secreted by adipose tissue and insulin resistance has been confirmed in human individuals, independently of obesity.[3] Raised leptin and insulin levels, the hallmarks of obesity-related insulin resistance, drive SPARC secretion from adipose tissue *in vitro*.[3]

SPARC has also been associated with diabetic retinopathy, as this protein is differentially expressed and secreted by retinal endothelial cells of individuals with and without T2DM.[80] SPARC is also expressed and secreted by retinal pigment basal membrane cells,[81] which show basal membrane thickening and permeability changes in patients with T2DM.[82,83] Furthermore, the expression of *SPARC* was increased in the vitreous body of patients with proliferative diabetic retinopathy and, in rats, subretinal injection of a recombinant adenovirus expressing *SPARC* increased preretinal neovascularisation in rats, which was further enhanced by addition of VEGF.[84]

Sparc-knockout mice develop senile cataracts and have impaired wound healing, two conditions that may complicate poor glycemic control in T2DM.[85,86] Although glucose seems to reduce SPARC levels *in vitro*,[3,26] whether these pathologies are triggered by a reduction of SPARC concentrations caused by hyperglycemia has not been established.

In addition, SPARC has been implicated in the pathogenesis of diabetic nephropathy, in which the basal membrane is typically thickened and the synthesis of ECM components increased. Patients with diabetic nephropathy have elevated circulating levels of SPARC.[87] Furthermore, histological changes associated with nephropathy are ameliorated in *Sparc*-knockout streptozotocin-induced diabetic mice compared with those in wild-type streptozotocin-induced diabetic mice; these *Sparc*-knockout mice had reduced ECM expansion around the mesangial cells of the kidney, with diminished accumulation of type IV collagen and laminin, and reduced accumulation of type I and type IV collagen in the ECM of the interstitial tubules.[88] Expression of the gene that encodes TGF-β1, which is one of the growth factors regulated by SPARC, in renal tissue was also reduced in these mice.[88] Human studies in individuals who underwent renal biopsies for diagnostic

reasons have shown that serum SPARC levels in individuals with T2DM were higher than those in controls and that SPARC correlated with severity of glomerular diffuse lesions, and with urinary albumin excretion and serum creatinine concentrations.[87,89]

SPARC is also expressed in the acinar cells of the pancreas in rats; gene expression is increased with inflammation (pancreatitis) and lost with destruction of acinar cells.[90] SPARC has been found in the same type of cells in human patients with chronic pancreatitis,[90] but no studies have been performed to determine whether this expression is associated with impairment of insulin secretion. Of note, T2DM is associated with pancreatic fibrosis, which is characterized by deposition of organized fibril-forming collagen and islet amyloid, and by adipocyte infiltration.[91,92] Whether SPARC plays a part in facilitating the fibrotic processes in a hyperglycemic setting is not known, although several mechanisms have been postulated, including upregulation of metalloproteinases, production of reactive oxygen species and mineralocorticoid activation, and this might be an important area for future research.

SPARC and cardiovascular disease

SPARC is essential to the regulation of the collagenous ECM of the heart, which is characterized by more insoluble fibril-forming collagen with a higher rate of turnover than that of other tissues.[93] In response to experimental myocardial infarction, *Sparc*-knockout mice experience increased cardiac rupture and cardiac-related mortality and have disorganized granulation tissue and immature collagenous ECM at the infarct site.[94] By contrast, overexpression of *SPARC* in wild-type mice improved collagen maturation after experimental myocardial infarction and prevented cardiac dilatation.[94] In addition, experimentally induced cardiac pressure overload in mice (which mimics diastolic dysfunction, a typical feature of human obesity-related heart disease)[95] is associated with decreased levels of fibril-forming collagen in *Sparc*-knockout animals compared with those in wild-type mice.[96]

Individuals with obesity have a higher risk of ischemic heart disease. In support of a role of SPARC in the pathogenesis of this disease, β-adrenergic stimulation increases *SPARC* expression and collagen content in rat hearts after exposure to isoprenaline (a β-adrenergic agonist, also known as isoproterenol).[63] Furthermore, elevated plasma SPARC concentrations have been found in human patients with existing ischemic cardiovascular disease.[4] In pig-tailed macaque (*Macaca nemestrina*), the expression of *SPARC* and platelet-derived growth factor (*PDGF*) genes in endothelial cells and vascular smooth muscle cells is enhanced in advanced atherosclerotic lesions.[97] Furthermore, SPARC regulates the binding of PDGF to its receptor in human fibroblasts *in vitro*.[97] Further research is required to establish whether elevated circulating SPARC, as found in obesity, damages the heart. A review of the current understanding of the relationship between SPARC and the heart has been published this year.[93]

Other obesity-related complications

Obesity is associated with increased incidence of cancer.[33] Increased SPARC gene and protein expression have been associated with worse prognosis of melanomas, meningiomas and breast cancer by promoting invasion and metastasis.[98] SPARC may inhibit the progression of tumors such as ovarian cancer.[99] The role of SPARC in cancer in the context of obesity is unclear, although this factor seems to influence principally tumor progression as opposed to tumor formation.[100]

Obesity is also associated with an increased risk of Alzheimer disease. Overexpression of SPARC in the brain might be involved in the pathogenesis of Alzheimer-associated neuronal atrophy by increasing neural cell death by detachment from the ECM.[101] SPARC gene and protein expression in the vascular basement membrane were reduced in rats following transient global brain ischemia, 24 h after cerebral reperfusion,[64] and this decrease in SPARC levels may have implications in promoting a blood–brain barrier breakdown after ischemic injury. The post-ischemic downregulation of SPARC was reduced by placing the animals in hypothermic conditions, which was also associated with reduced blood–brain barrier breakdown.[64] The exact role of SPARC in cerebrovascular disease remains, however, unclear.

Obesity is a condition characterized by subinflammatory changes. Besides the induction of SPARC by inflammatory stimuli, SPARC's role in initiation and maintenance of inflammatory conditions is yet poorly defined. SPARC binds to vascular cell adhesion protein 1 (also known as VCAM1) to enable transendothelial migration, and *Sparc* knock out substantially impairs migration of leukocytes and neutrophils.[102] Increased SPARC levels have been identified in the synovial fluid of patients with rheumatoid arthritis, a condition in which levels of the proinflammatory cytokine leptin are also elevated; the increase in SPARC may result from leptin stimulation.[103]

SPARC and the skeletal system

Individuals with obesity have an increased bone mass. An increased mechanical loading by total body mass is currently thought to be the mechanism by which bone is strengthened in obesity.[104] SPARC, which is secreted by osteoblasts into osteoid, may, however, also have a protective role against osteoporosis in these patients. *Sparc*-knockout mice have low-turnover osteopenia, which suggests that SPARC may strengthen bone. This effect probably occurs by SPARC binding to integrin receptors, thus modulating Wnt/β-catenin signaling, which leads to decreased adipogenesis and increased osteogenesis.[24,58] A study on *Sparc*-null osteoblasts has suggested that SPARC may also inhibit the Notch signaling pathway and lead to enhanced osteoblast differentiation.[105]

In contrast to the mechanisms for the potential protective effect of SPARC in bone remodeling, whether SPARC has protective effects on cartilage is less clear. Increased levels of the SPARC protein are found in cartilage of individuals with osteoarthritis or rheumatoid arthritis.[67] Achondroplasia is associated with a protein

trafficking deficit, to which retention of SPARC in the endoplasmatic reticulum of chondrocytes may contribute, and accelerated vertebral disc degeneration is found in *Sparc*-knockout mice.[106,107] Therefore, a careful balance in the secretion of SPARC may be necessary for normal physiology. Of note, leptin, which is increasingly recognized as an important regulator of bone homeostasis and has proinflammatory potential, may be responsible for the raise in SPARC secretion that is observed in inflammatory conditions associated with the progression of arthritis in obesity.[46,108]

SPARC and aging

ECM composition changes with age; collagen degradation is reduced by a decrease in the concentration of matrix metalloproteinases and collagen accumulation is augmented by an increase in the concentration of inhibitors of matrix metalloproteinases.[109] Some age-related pathologies might be explained by the effects of SPARC on collagen fibril assembly and procollagen processing.[110] SPARC is expressed at higher levels during development and its secretion seems to decline progressively throughout adult life.[111] Many phenotypic changes observed in *Sparc*-knockout mice, such as accelerated vertebral disc degeneration, osteoporosis and the development of cataracts mimic age-related changes. In addition, a reduced expression of *SPARC* was shown in older donors of fibroblasts and endothelial cells in mice and humans, but, in mouse cells, the effect of age on changes in collagen composition and reduced VEGF levels was reversed in part by exposure of primary-cell fibroblast cultures to TGF-β1.[112] These findings are in contradiction with the observation of increased *SPARC* expression in patients with Werner syndrome, which is characterized by premature aging,[113] and further studies are necessary to examine whether the expression of SPARC in individual organs is equally affected by aging.

Therapeutic manipulation of SPARC

Obesity-associated elevations of SPARC secretion may have adverse effects on normal physiology; reduction of SPARC levels or aspects of SPARC function may thus have therapeutic potential. Weight loss reduces SPARC expression,[3] but, as pharmacotherapy has limited efficacy in weight management, we expect that SPARC inhibition would not aid weight loss but could potentially ameliorate obesity-associated complications. SPARC suppression has previously been used in an experimental, *in vitro* model of cancer treatment: the nonsteroidal anti-inflammatory drug NS398, a selective inhibitor of prostaglandin G/H synthase 2 (also known as cyclooxygenase 2), suppressed *SPARC* expression in lung cancer cell lines.[114] The same drug may have benefits in the treatment of colon cancer and esophageal cancer, as it induced apoptosis in cell cancer lines,[115,116] but this treatment has not yet been used clinically. Furthermore, adenovirus-mediated inhibition of SPARC expression reduced experimentally induced liver fibrosis in rats.[117] Manipulation of SPARC may provide new therapeutic avenues in obesity and its associated complications, especially in the prevention and treatment of the microvascular and macrovascular complications of diabetes mellitus, but further research into the effects of SPARC inhibition in human physiology is necessary.

Conclusions

SPARC, an extracellular glycoprotein, is expressed ubiquitously, but predominantly in adipocytes. SPARC is a regulator of adipocyte differentiation and composition of the ECM which, in obesity, is characterized by increased collagen fibril content consistent with fibrosis. Fibrosis of adipose tissue limits its capacity for expansion and storage of triglycerides, which spread to the circulation and are stored in organs such as the liver, muscle and pancreas, and as perivascular adipose tissue. This ectopic storage of lipids is linked with insulin resistance and metabolic complications. Increased levels of SPARC are found in obesity and this increase is most likely due to increased SPARC secretion from adipose tissue. SPARC may contribute to the development of obesity-associated complications as a result of increased ectopic triglycerid content, which also links obesity to increased insulin resistance. As suggested by Naïmi and Van Obberghen,[69] the quality and expansibility of adipose tissue might be more important than adipose mass *per se*.

SPARC may independently contribute to the pathogenesis of diabetic nephropathy and retinopathy, owing to its profibrotic and angiogenic effects. Furthermore, SPARC may contribute to the pathogenesis of other obesity-associated disorders, including heart failure, some obesity-associated cancers, premature aging and osteoarthritis, whereas the elevated SPARC levels found in obesity may be beneficial to the bone by supporting its strength. Although suppression of SPARC could be a promising therapy for obesity, as it could protect individuals from the adverse effects of this condition, including development of T2DM, without affecting body weight *per se,* further research is necessary to determine its pharmacologic potential.

Review criteria

A literature search was performed on PubMed and MEDLINE for articles and abstracts published from January 1981 to November 2009. The search terms "SPARC", "osteonectin", "adipose tissue", "obesity", "insulin resistance" and "diabetes" were used to identify reviews. The search terms "cardiovascular disease", "diastolic dysfunction", "ischemic heart disease", "cerebrovascular disease", "stroke", "fatty liver disease", "steatohepatitis", "osteoarthritis", "atherosclerosis", "diabetic retinopathy", "cataract" and "diabetic nephropathy" were used in combination with "SPARC" and "osteonectin" to identify original research papers and were supplemented by manual search of bibliographies of pertinent articles. Conference abstracts were screened for further leads. English-language articles and abstracts, and a few abstracts of non-English-language articles, were considered.

1. Termine, J. D. *et al.* Osteonectin, a bone-specific protein linking mineral to collagen. *Cell* **26**, 99–105 (1981).
2. Schulz, A., Jundt, G., Berghäuser, K. H., Gehron-Robey, P. & Termine, J. D. Immunohistochemical study of osteonectin in various types of osteosarcoma. *Am. J. Pathol.* **132**, 233–238 (1988).
3. Kos, K. *et al.* Regulation of the fibrosis and angiogenesis promoter SPARC in human adipose tissue by weight change, leptin, insulin and glucose. *Diabetes* **58**, 1780–1788 (2009).
4. Takahashi, M. *et al.* The expression of SPARC in adipose tissue and its increased plasma concentration in patients with coronary artery disease. *Obes. Res.* **9**, 388–393 (2001).
5. Henegar, C. *et al.* Adipose tissue transcriptomic signature highlights the pathological relevance of extracellular matrix in human obesity. *Genome Biol.* **9**, R14 (2008).
6. Sage, E. H., Johnson, C. & Bornstein, P. Characterization of a novel serum albumin-binding glycoprotein secreted by endothelial cells in culture. *J. Biol. Chem.* **259**, 3993–4007 (1984).
7. Saltman, D. L., Dolganov, G. M., Warrington, J. A., Wasmuth, J. J. & Lovett, M. A physical map of 15 loci on human chromosome 5q23-q33 by two-color fluorescence *in situ* hybridization. *Genomics* **16**, 726–732 (1993).
8. Yan, Q. & Sage, E. H. SPARC, a matricellular glycoprotein with important biological functions. *J. Histochem. Cytochem.* **47**, 1495–1506 (1999).
9. Stenner, D. D. *et al.* Monoclonal antibodies to native noncollagenous bone-specific proteins. *Proc. Natl Acad. Sci. USA* **81**, 2868–2872 (1984).
10. Sage, E. H. & Vernon, R. B. Regulation of angiogenesis by extracellular matrix: the growth and the glue. *J. Hypertens.* **12** (Suppl.), S145–S152 (1995).
11. Sasaki, T. *et al.* Limited cleavage of extracellular matrix protein BM-40 by matrix metalloproteinases increases its affinity for collagens. *J. Biol. Chem.* **272**, 9237–9243 (1997).
12. Sasaki, T., Hohenester, E., Göhring, W. & Timpl, R. Crystal structure and mapping by site-directed mutagenesis of the collagen-binding epitope of an activated form of BM-40/SPARC/osteonectin. *EMBO J.* **17**, 1625–1634 (1998).
13. Bornstein, P. Diversity of function is inherent in matricellular proteins: an appraisal of thrombospondin 1. *J. Cell Biol.* **130**, 503–506 (1995).
14. Brekken, R. A. & Sage, E. H. SPARC, a matricellular protein: at the crossroads of cell-matrix communication. *Matrix Biol.* **19**, 816–827 (2001).
15. Kupprion, C., Motamed, K. & Sage, E. H. SPARC (BM-40, osteonectin) inhibits the mitogenic effect of vascular endothelial growth factor on microvascular endothelial cells. *J. Biol. Chem.* **273**, 29635–29640 (1998).
16. Ruhrberg, C. Growing and shaping the vascular tree: Multiple roles for VEGF. *Bioessays* **25**, 1052–1060 (2003).
17. Francki, A. & Sage, E. H. SPARC and the kidney glomerulus: matricellular proteins exhibit diverse functions under normal and pathological conditions. *Trends Cardiovasc. Med.* **11**, 32–37 (2001).
18. Long, M. W. Osteogenesis and bone-marrow-derived cells. *Blood Cells Mol. Dis.* **27**, 677–690 (2001).
19. Jørgensen, L. H. *et al.* Secreted protein acidic and rich in cysteine (SPARC) in human skeletal muscle. *J. Histochem. Cytochem.* **57**, 29–39 (2009).
20. Tartare-Deckert, S., Chavey, C., Monthouel, M. N., Gautier, N. & Van Obberghen, E. The matricellular protein SPARC/osteonectin as a newly identified factor up-regulated in obesity. *J. Biol. Chem.* **276**, 22231–22237 (2001).
21. Clark, C. J. & Sage, E. H. A prototypic matricellular protein in the tumor microenvironment—where there's SPARC, there's fire. *J. Cell. Biochem.* **104**, 721–732 (2008).
22. Nie, J. *et al.* IFATS collection: Combinatorial peptides identify alpha5beta1 integrin as a receptor for the matricellular protein SPARC on adipose stromal cells. *Stem Cells* **26**, 2735–2745 (2008).
23. Kelm, R. J. Jr, Swords, N. A., Orfeo, T. & Mann, K. G. Osteonectin in matrix remodeling. A plasminogen-osteonectin-collagen complex. *J. Biol. Chem.* **269**, 30147–30153 (1994).
24. Barker, T. H. *et al.* Matricellular homologs in the foreign body response: hevin suppresses inflammation, but hevin and SPARC together diminish angiogenesis. *Am. J. Pathol.* **166**, 923–933 (2005).
25. Delany, A. M. *et al.* Osteopenia and decreased bone formation in osteonectin-deficient mice. *J. Clin. Invest.* **105**, 915–923 (2000).
26. Mansergh, F. C. *et al.* Osteopenia in Sparc (osteonectin)-deficient mice: characterization of phenotypic determinants of femoral strength and changes in gene expression. *Physiol. Genomics* **32**, 64–73 (2007).
27. Gilmour, D. T. *et al.* Mice deficient for the secreted glycoprotein SPARC/osteonectin/BM40 develop normally but show severe age-onset cataract formation and disruption of the lens. *EMBO J.* **17**, 1860–1870 (1998).
28. Bradshaw, A. D. *et al.* SPARC-null mice display abnormalities in the dermis characterized by decreased collagen fibril diameter and reduced tensile strength. *J. Invest. Dermatol.* **120**, 949–955 (2003).
29. Bradshaw, A. D., Graves, D. C., Motamed, K. & Sage, E. H. SPARC-null mice exhibit increased adiposity without significant differences in overall body weight. *Proc. Natl Acad. Sci. USA* **100**, 6045–6050 (2003).
30. Colditz, G. A., Willett, W. C., Rotnitzky, A. & Manson, J. E. Weight gain as a risk factor for clinical diabetes in women. *Ann. Intern. Med.* **122**, 481–486 (1995).
31. International Diabetes Federation. *IDF Consensus Worldwide Definition of the Metabolic Syndrome* [online], www.idf.org/webdata/docs/IDF_Meta_def_final.pdf (2006).
32. Poirier, P. *et al.* Obesity and cardiovascular disease: pathophysiology, evaluation, and effect of weight loss: an update of the 1997 American Heart Association Scientific Statement on Obesity and Heart Disease from the Obesity Committee of the Council on Nutrition, Physical Activity, and Metabolism. *Circulation* **113**, 898–918 (2006).
33. Renehan, A. G., Tyson, M., Egger, M., Heller, R. F. & Zwahlen, M. Body-mass index and incidence of cancer: a systematic review and meta-analysis of prospective observational studies. *Lancet* **371**, 569–578 (2008).
34. Chun, T. H. *et al.* A pericellular collagenase directs the 3-dimensional development of white adipose tissue. *Cell* **125**, 577–591 (2006).
35. Khan, T. *et al.* Metabolic dysregulation and adipose tissue fibrosis: the role of collagen VI. *Mol. Cell. Biol.* **29**, 1575–1591 (2009).
36. Danforth, E. Jr. Failure of adipocyte differentiation causes type II diabetes mellitus? *Nat. Genet.* **26**, 13 (2000).
37. Lee, D. E., Kehlenbrink, S., Lee, H., Hawkins, M. A. & Yudkin, J. S. Getting the message across: mechanisms of physiological cross-talk by adipose tissue. *Am. J. Physiol. Endocrinol. Metab.* **296**, E1210–E1229 (2009).
38. Wong, S. P. *et al.* Adipokines and the insulin resistance syndrome in familial partial lipodystrophy caused by a mutation in lamin A/C. *Diabetologia* **48**, 2641–2649 (2005).
39. Greenstein, A. S. *et al.* Local inflammation and hypoxia abolish the protective anticontractile properties of perivascular fat in obese patients. *Circulation* **119**, 1661–1670 (2009).
40. Guzik, T. J., Marvar, P. J., Czesnikiewicz-Guzik, M. & Korbut, R. Perivascular adipose tissue as a messenger of the brain-vessel axis: role in vascular inflammation and dysfunction. *J. Physiol. Pharmacol.* **58**, 591–610 (2007).
41. Kim, J. Y. *et al.* Obesity-associated improvements in metabolic profile through expansion of adipose tissue. *J. Clin. Invest.* **117**, 2621–2637 (2007).
42. Wong, S. P. *et al.* Adipokines and the insulin resistance syndrome in familial partial lipodystrophy caused by a mutation in lamin A/C. *Diabetologia* **48**, 2641–2649 (2005).
43. Ravussin, E. & Smith, S. R. Increased fat intake, impaired fat oxidation, and failure of fat cell proliferation result in ectopic fat storage, insulin resistance, and type 2 diabetes mellitus. *Ann. NY Acad. Sci.* **967**, 363–378 (2002).
44. Després, J. P. & Lemieux, I. Abdominal obesity and metabolic syndrome. *Nature* **444**, 881–887 (2006).
45. Trayhurn, P. & Beattie, J. H. Physiological role of adipose tissue: white adipose tissue as an endocrine and secretory organ. *Proc. Nutr. Soc.* **60**, 329–339 (2001).
46. Kos, K. & Wilding, J. P. Adipokines: Emerging therapeutic targets. *Curr. Opin. Investig. Drugs* **10**, 1061–1068 (2009).
47. Berg, A. & Scherer, P. E. Adipose tissue, inflammation and cardiovascular risk. *Circ. Res.* **96**, 939–949 (2005).
48. Fantuzzi, G. & Mazzone, T. Adipose tissue and atherosclerosis: exploring the connection. *Ateriosclr. Thromb. Vasc. Biol.* **27**, 996–1003 (2007).
49. Keophiphath, M. *et al.* Macrophage-secreted factors promote a profibrotic phenotype in human preadipocytes. *Mol. Endocrinol.* **23**, 11–24 (2009).
50. Skurk, T. *et al.* Production and release of macrophage migration inhibitory factor from human adipocytes. *Endocrinology* **146**, 1006–1011 (2005).
51. Halberg, N. *et al.* Hypoxia-inducible factor 1alpha induces fibrosis and insulin resistance in white adipose tissue. *Mol. Cell. Biol.* **29**, 4467–4483 (2009).
52. Trayhurn, P. & Wood, I. S. Adipokines: inflammation and the pleiotropic role of white adipose tissue. *Br. J. Nutr.* **92**, 347–355 (2004).
53. Ye, J., Gao, Z., Yin, J. & He, Q. Hypoxia is a potential risk factor for chronic inflammation and adiponectin reduction in adipose tissue of ob/ob and dietary obese mice. *Am. J. Physiol. Endocrinol. Metab.* **293**, E1118–E1128 (2007).
54. Pasarica, M. *et al.* Reduced adipose tissue oxygenation in human obesity: evidence for rarefaction, macrophage chemotaxis, and inflammation without an angiogenic response. *Diabetes* **58**, 718–725 (2009).
55. Wang, B., Wood, I. S. & Trayhurn, P. Dysregulation of the expression and secretion of inflammation-related adipokines by hypoxia in human adipocytes. *Pflugers Arch.* **455**, 479–492 (2007).

56. Higami, Y. *et al.* Energy restriction lowers the expression of genes linked to inflammation, the cytoskeleton, the extracellular matrix, and angiogenesis in mouse adipose tissue. *J. Nutr.* **136**, 343–352 (2006).
57. Chavey, C. *et al.* Regulation of secreted protein acidic and rich in cysteine during adipose conversion and adipose tissue hyperplasia. *Obesity* **14**, 1890–1897 (2006).
58. Nie, J. & Sage, E. H. SPARC inhibits adipogenesis by its enhancement of beta-catenin signaling. *J. Biol. Chem.* **284**, 1279–1290 (2009).
59. O'Connor, K. C., Song, H., Rosenzweig, N. & Jansen, D. A. Extracellular matrix substrata alter adipocyte yield and lipogenesis in primary cultures of stromal-vascular cells from human adipose. *Biotechnol. Lett.* **25**, 1967–1972 (2003).
60. Barker, T. H. *et al.* SPARC regulates extracellular matrix organization through its modulation of integrin-linked kinase activity. *J. Biol. Chem.* **280**, 36483–36493 (2005).
61. Fliers, E. *et al.* White adipose tissue: getting nervous. *J. Neuroendocrinol*. **15**, 1005–1010 (2003).
62. Ricci, M. R. *et al.* Isoproterenol decreases leptin release from rat and human adipose tissue through posttranscriptional mechanisms. *Am. J. Physiol. Endocrinol. Metab.* **288**, E798–E804 (2005).
63. Masson, S. *et al.* Remodelling of cardiac extracellular matrix during beta-adrenergic stimulation: upregulation of SPARC in the myocardium of adult rats. *J. Mol. Cell. Cardiol.* **30**, 1505–1514 (1998).
64. Baumann, E., Preston, E., Slinn, J. & Stanimirovic, D. Post-ischemic hypothermia attenuates loss of the vascular basement membrane proteins, agrin and SPARC, and the blood-brain barrier disruption after global cerebral ischemia. *Brain Res*. **1269**, 185–197 (2009).
65. Vincent, A. J., Lau, P. W. & Roskams, A. J. SPARC is expressed by macroglia and microglia in the developing and mature nervous system. *Dev. Dyn.* **237**, 1449–1462 (2008).
66. Wu, R. X. *et al.* Fibroblast migration after myocardial infarction is regulated by transient SPARC expression. *J. Mol. Med.* **84**, 241–252 (2006).
67. Nakamura, S. *et al.* Enhancement of SPARC (osteonectin) synthesis in arthritic cartilage Increased levels in synovial fluids from patients with rheumatoid arthritis and regulation by growth factors and cytokines in chondrocyte cultures. *Arthritis Rheum.* **39**, 539–551 (1996).
68. Kzhyshkowska, J. *et al.* Novel function of alternatively activated macrophages: stabilin-1-mediated clearance of SPARC. *J. Immunol.* **176**, 5825–5832 (2006).
69. Naïmi, M. & Van Obberghen, E. Inflammation: where is the SPARC in adipose-tissue inflammation? *Nat. Rev. Endocrinol.* **5**, 648–649 (2009).
70. Koukourakis, M. I. *et al.* Enhanced expression of SPARC/osteonectin in the tumor-associated stroma of non-small cell lung cancer is correlated with markers of hypoxia/acidity and with poor prognosis of patients. *Cancer Res.* **63**, 5376–5380 (2003).
71. Wang, B., Wood, I. S. & Trayhurn, P. Hypoxia induces leptin gene expression and secretion in human preadipocytes: differential effects of hypoxia on adipokine expression in preadipocytes. *J. Endocrinol.* **198**, 127–134 (2008).
72. Chlenski A. *et al.* SPARC expression is associated with impaired tumor growth, inhibited angiogenesis and changes in the extracellular matrix. *Int. J. Cancer.* **118**, 310–316 (2006).
73. Sage, E. H. *et al.* Cleavage of the matricellular protein SPARC by matrix metalloproteinase 3 produces polypeptides that influence angiogenesis. *J. Biol. Chem.* **278**, 37849–37857 (2003).
74. Fain, J. N., Tichansky, D. S. & Madan, A. K. Transforming growth factor β1 release by human adipose tissue is enhanced in obesity. *Metabolism* **54**, 1546–1551 (2005).
75. Wolf, G. *et al.* Leptin stimulates proliferation and TGF-beta expression in renal glomerular endothelial cells: potential role in glomerulosclerosis. *Kidney Int.* **56**, 860–872 (1999).
76. Kao, Y. H. *et al.* Serum factors potentiate hypoxia-induced hepatotoxicity *in vitro* through increasing transforming growth factor-beta1 activation and release. *Cytokine* **47**, 11–22 (2009).
77. Higgins, D. F., Kimura, K., Iwano, M. & Haase, V. H. Hypoxia-inducible factor signaling in the development of tissue fibrosis. *Cell Cycle* **7**, 1128–1132 (2008).
78. Oltmanns, K. M. *et al.* Hypoxia causes glucose intolerance in humans. *Am. J. Respir. Crit. Care Med.* **169**, 1231–1237 (2004).
79. Jakobsson, P. & Jorfeldt, L. Oxygen supplementation increases glucose tolerance during euglycaemic hyperinsulinaemic glucose clamp procedure in patients with severe COPD and chronic hypoxaemia. *Clin. Physiol. Funct. Imaging* **26**, 271–274 (2006).
80. Munjal, I. D., McLean, N. V., Grant, M. B. & Blake, D. A. Differences in the synthesis of secreted proteins in human retinal endothelial cells of diabetic and nondiabetic origin. *Curr. Eye Res.* **13**, 303–310 (1994).
81. Ratnayaka, A. *et al.* Trafficking of osteonectin by retinal pigment epithelial cells: evidence for basolateral secretion. *Int. J. Biochem. Cell Biol.* **39**, 85–92 (2007).
82. Grimes, P. A., McGlinn, A., Laties, A. M. & Naji, A. Increase of basal cell membrane area of the retinal pigment epithelium in experimental diabetes. *Exp. Eye Res.* **38**, 569–577 (1984).
83. Chakrabarti, S., Prashar, S. & Sima, A. A. Augmented polyol pathway activity and retinal pigment epithelial permeability in the diabetic BB rat. *Diabetes Res. Clin. Pract.* **8**, 1–11 (1990).
84. Watanabe, K. *et al.* SPARC is a major secretory gene expressed and involved in the development of proliferative diabetic retinopathy. *J. Atheroscler. Thromb.* **16**, 69–76 (2009).
85. Rowe, N. G., Mitchell, P. G., Cumming, R. G. & Wans, J. J. Diabetes, fasting blood glucose and age-related cataract: the Blue Mountains Eye Study. *Ophthalmic Epidemiol.* **7**, 103–114 (2000).
86. Yue, D. K. *et al.* Effects of experimental diabetes, uremia, and malnutrition on wound healing. *Diabetes* **36**, 295–299 (1987).
87. Kanauchi, M., Nishioka, M. & Dohi, K. Secreted protein acidic and rich in cysteine (SPARC) in patients with diabetic nephropathy and tubulointerstitial injury. *Diabetologia* **43**, 1076–1077 (2000).
88. Taneda, S. *et al.* Amelioration of diabetic nephropathy in SPARC-null mice. *J. Am. Soc. Nephrol.* **14**, 968–980 (2003).
89. Kanauchi, M., Nishioka, H., Kawano, T. & Dohi, K. Role of secreted protein acidic and rich in cysteine (SPARC) in patients with diabetic nephropathy. *Clin. Exper. Nephrol.* **1**, 115–120 (1997).
90. Reding, T. *et al.* Inflammation-dependent expression of SPARC during development of chronic pancreatitis in WBN/Kob rats and a microarray gene expression analysis. *Physiol. Genomics* **38**, 196–204 (2009).
91. Clark, A. *et al.* Islet amyloid, increased A-cells, reduced B-cells and exocrine fibrosis: quantitative changes in the pancreas in type 2 diabetes. *Diabetes Res.* **9**, 151–159 (1988).
92. Hayden, M. R. *et al.* Attenuation of endocrine-exocrine pancreatic communication in type 2 diabetes: pancreatic extracellular matrix ultrastructural abnormalities. *J. Cardiometab. Syndr.* **3**, 234–243 (2008).
93. McCurdy, S., Baicu, C. F., Heymans, S. & Bradshaw, A. D. Cardiac extracellular matrix remodeling: Fibrillar collagens and Secreted Protein Acidic and Rich in Cysteine (SPARC). *J. Mol. Cell Cardiol.* doi:10.1016/j.yjmcc.2009.06.018.
94. Schellings, M. W. *et al.* Absence of SPARC results in increased cardiac rupture and dysfunction after acute myocardial infarction. *J. Exp. Med.* **206**, 113–123 (2009).
95. Powell, B. D., Redfield, M. M., Bybee, K. A., Freeman, W. K. & Rihal, C. S. Association of obesity with left ventricular remodeling and diastolic dysfunction in patients without coronary artery disease. *Am. J. Cardiol.* **98**, 116–120 (2006).
96. Bradshaw, A. D. *et al.* Pressure overload-induced alterations in fibrillar collagen content and myocardial diastolic function: role of secreted protein acidic and rich in cysteine (SPARC) in post-synthetic procollagen processing. *Circulation* **119**, 269–280 (2009).
97. Raines, E. W., Lane, T. F., Iruela-Arispe, M. L., Ross, R. & Saga, E. H. The extracellular glycoprotein SPARC interacts with platelet derived growth factor (PDGF)-AB and -BB and inhibits the binding of PDGF to its receptors. *Proc. Natl Acad. Sci. USA* **89**, 1281–1285 (1992).
98. Sangaletti, S. *et al.* Macrophage-derived SPARC bridges tumor cell-extracellular matrix interactions toward metastasis. *Cancer Res.* **68**, 9050–9059 (2008).
99. Said, N. & Motamed, K. Absence of host-secreted protein acidic and rich in cysteine (SPARC) augments peritoneal ovarian carcinomatosis. *Am. J. Pathol*. **167**, 1739–1752 (2005).
100. Podhajcer, O. L. *et al.* The role of the matricellular protein SPARC in the dynamic interaction between the tumor and the host. *Cancer Metastasis Rev.* **27**, 523–537 (2008).
101. Kögel, D., Schomburg, R., Copanaki, E. & Prehn, J. H. Regulation of gene expression by the amyloid precursor protein: inhibition of the JNK/c-Jun pathway. *Cell Death Differ.* **12**, 1–9 (2005).
102. Kelly, K. A. *et al.* SPARC is a VCAM-1 counter-ligand that mediates leukocyte transmigration. *J. Leukc. Biol*. **81**, 748–756 (2007).
103. Lago, R., Gómez, R., Lago, F., Gómez-Reino, J. & Gualillo, O. Leptin beyond body weight regulation-current concepts concerning its role in immune function and inflammation. *Cell. Immunol.* **252**, 139–145 (2008).
104. Zhao, L. J. *et al.* Relationship of obesity with osteoporosis. *J. Clin. Endocrinol. Metab.* **92**, 1640–1646 (2007).
105. Kessler, C. B. & Delany, A. M. Increased Notch 1 expression and attenuated stimulatory G protein coupling to adenylyl cyclase in osteonectin-null osteoblasts. *Endocrinology* **148**, 1666–1674 (2007).

106. Hecht, J. T. & Sage, E. H. Retention of the matricellular protein SPARC in the endoplasmic reticulum of chondrocytes from patients with pseudoachondroplasia. *J. Histochem. Cytochem.* **54**, 269–274 (2006).
107. Gruber, H. E. *et al.* Targeted deletion of the SPARC gene accelerates disc degeneration in the aging mouse. *J. Histochem. Cytochem*. **53**, 1131–1138 (2005).
108. Shi, Y. *et al.* Dissociation of the neuronal regulation of bone mass and energy metabolism by leptin *in vivo*. *Proc. Natl Acad. Sci. USA* **105**, 20529–20533 (2008).
109. Lindsey, M. L. *et al.* Age-dependent changes in myocardial matrix metalloproteinase profiles and fibroblast function. *Cardiovasc. Res.* **66**, 410–419 (2005).
110. Bradshaw, A. D. & Sage, E. H. SPARC, a matricellular protein that functions in cellular differentiation and tissue response to injury. *J. Clin. Invest.* **107**, 1049–1054 (2001).
111. Lane, T. F. & Sage, E. H. The biology of SPARC, a protein that modulates cell-matrix interactions. *FASEB J.* **8**, 163–173 (1994).
112. Reed, M. J. *et al.* Enhanced angiogenesis characteristic of SPARC-null mice disappears with age. *J. Cell Physiol.* **204**, 800–807 (2005).
113. Lecka-Czernik, B., Moerman, E. J., Jones, R. A. & Goldstein, S. Identification of gene sequences overexpressed in senescent and Werner syndrome human fibroblasts. *Exp. Gerontol*. **31**, 159–174 (1996).
114. Pan, M. R., Chang, H. C., Chuang, L. Y. & Hung, W. C. The nonsteroidal anti-inflammatory drug NS398 reactivates SPARC expression via promoter demethylation to attenuate invasiveness of lung cancer cells. *Exp. Biol. Med.* **233**, 456–462 (2008).
115. Li, M., Wu, X. & Xu, X. C. Induction of apoptosis in colon cancer cells by cyclooxygenase-2 inhibitor NS398 through a cytochrome c-dependent pathway. *Clin. Cancer Res*. **7**, 1010–1016 (2001).
116. Li, M., Wu, X. & Xu, X. C. Induction of apoptosis by cyclo-oxygenase-2 inhibitor NS398 through a cytochrome C-dependent pathway in esophageal cancer cells. *Int. J. Cancer* **93**, 218–223 (2001).
117. Camino, A. M. *et al.* Adenovirus-mediated inhibition of SPARC attenuates liver fibrosis in rats. *J. Gene Med.* **10**, 993–1004 (2008).