Immune regulators of inflammation in obesity-associated type 2 diabetes and coronary artery disease

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Purpose of review
To summarize current work identifying inflammatory components that underlie associations between obesity-associated type 2 diabetes and coronary artery disease.

Recent findings
Recent studies implicate immune cells as drivers of pathogenic inflammation in human type 2 diabetes. Inflammatory lymphocytes characterize unhealthy adipose tissue, but regional adipose volume, primarily visceral and pericardial fat, also predict severity and risk for obesity-associated coronary artery disease. Having a greater understanding of shared characteristics between inflammatory cells from different adipose tissue depots and a more accessible tissue, such as blood, will facilitate progress toward clinical translation of our appreciation of obesity as an inflammatory disease.

Summary
Obesity predisposes inflammation and metabolic dysfunction through multiple mechanisms, but these mechanisms remain understudied in humans. Studies of obese patients have identified disproportionate impacts of specific T cell subsets in metabolic diseases like type 2 diabetes. On the basis of demonstration that adipose tissue inflammation is depot-specific, analysis of adiposity by waist-to-hip ratio or MRI will increase interpretive value of lymphocyte-focused studies and aid clinicians in determining which obese individuals are at highest risk for coronary artery disease. New tools to combat obesity-associated coronary artery disease and other comorbidities will stem from identification of immune cell-mediated inflammatory networks that are amenable to pharmacological interventions.

Keywords
B cells, coronary artery disease, inflammation, obesity, T cell, type 2 diabetes

INTRODUCTION

The prevalence of obesity continues to increase worldwide and with current trajectories obesity will burden healthcare systems for decades [1]. Obesity often triggers the inflammation that is believed to increase risk for many comorbidities, with greatest risk stemming from the simmering inflammation that underlies type 2 diabetes (T2D) and coronary artery disease (CAD) [2▪,3▪▪]. Obesity-associated inflammation is largely due to overproduction of proinflammatory cytokines by macrophages [4,5], B cells and T cells [6–9,10▪▪], all of which are recruited to expanding adipose tissue. Immune cell-mediated inflammation reinforces a proinflammatory balance of adipokines, or adipocyte cytokines, which are also increased in expanding adipose tissue [11]. This combination of cytokines from adipose-associated immune cells and adipokines promotes metabolic dysregulation that includes insulin resistance and T2D [12▪,13]. The proinflammatory cytokine profile in obesity likely spills over into the circulation and predicts increased risk for CAD [14,15], pulmonary diseases [16–18] and cancer [19]. Inflammation may thereby link seemingly disparate comorbidities of obesity.

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KEY POINTS

- Obesity/insulin resistance causes a shift towards proinflammatory T helper 17 immune response in humans, which differs from the T helper 1/CD8$^+$ T cell dominance of murine obesity/insulin resistance.
- Proinflammatory T cell responses may underlie increased risk for obesity-associated CAD.
- Classifying patients within obese cohorts by depot-specific immune responses or by specific adipose depot volume will assist in developing criteria for obesity-associated inflammation, CAD risk and severity.

Although mouse models have enabled substantial progress over the last decade in describing immune compartment contributions to metabolic dysfunction, an understanding of the role inflammation plays in human obesity and associated metabolic disease remains rudimentary. Recent findings from human studies reviewed herein highlight ongoing progress in our understanding of inflammation as a driver of obesity-associated disease with emphasis on CAD.

OBESITY-ASSOCIATED INFLAMMATION AND IMMUNE CELLS IN TYPE 2 DIABETES

A major advance in metabolic research came with the understanding of obesity as a chronic, low-grade inflammatory state, which differs from the immune response to infection but involves well understood modulators of nutrient storage and efflux pathways [8,20,21▪]. Observations that activated macrophages within adipose tissue produce the majority of adipose-associated proinflammatory cytokines, and these cytokines promote insulin resistance [4,5], provoked the current focus on multiple types of immune cells in metabolic research and founded the field of 'Immunometabolism'. Extensive genetic, dietary and pharmacologic interventions in mouse models in parallel with observations from human studies identified a causal link between obesity-associated inflammation and metabolic disease [22,23]. Such studies also revealed a role for macrophages in remodeling within adipose tissue of lean individuals that contrasts with proinflammatory functions of newly recruited and/or in-situ proliferating macrophages of obese adipose tissue [24▪]. Other myeloid immune cells including neutrophils, eosinophils and mast cells also play roles in promoting inflammatory responses and insulin resistance in obese adipose tissue [25–27].

Classically designated 'adaptive' immune cells, including B cells, CD8$^+$ and CD4$^+$ T cells, also infiltrate adipose tissue and increase in number in response to obesity [28▪,29] (Fig. 1). Both B and T cells release and/or stimulate release of proinflammatory cytokines in obesity-associated insulin resistance/T2D, bolstering local and systemic inflammation. In addition to the B cell-intrinsic changes in T2D humans and obese/insulin resistance mice, B cells stimulate inflammatory cytokine production by CD4$^+$ and CD8$^+$ T cells [6,10▪▪]. Thus, B cells promote T2D-associated inflammation through direct (cell-intrinsic) and indirect (T cell-mediated) mechanisms. Further work with human samples showed that contact between B and T cells is required for maximal proinflammatory CD4$^+$ T helper 17 cell function in samples from T2D but not from obese, nondiabetic subjects. Although anti-inflammatory functions of B cells and regulatory T cells (Tregs) have also been well defined, these functions appear to be diminished in obesity [8,30–32]. For example, B cells from lean, 'metabolically healthy' humans and mice release significant amounts of the anti-inflammatory cytokine interleukin-10, but B cell interleukin-10 is severely downregulated in response to obesity/insulin resistance/T2D [10▪▪,33]. This shift to a potentially pathogenic, proinflammatory B cell cytokine profile may mechanistically underlie demonstrations that B cell-null mice fed a high-fat diet (HFD) are equally obese but less prone to obesity-associated insulin resistance and other metabolic disturbances. Anti-inflammatory CD4$^+$ Tregs are similarly underrepresented in obese/insulin resistant mice and T2D humans, in part because of lower numbers of Tregs [8,34] but also likely due to suppression of anti-inflammatory interleukin-10 production [35▪], although clarification is needed in follow-up studies with human samples. Despite overall similarities in roles for B and T cells in human and mouse T2D, a more comprehensive assessment of T cells in obesity suggests fundamental differences in obesity-associated inflammation between humans and mice: obese mice have predominantly CD8 and T helper 1 inflammatory responses with minor changes in T helper 17 cells [8,9], whereas in obese humans, the T helper 17 axis dominates T cell-mediated inflammation [10▪▪,33,36,37▪▪].

T HELPER 17 CELLS IN OBESITY AND TYPE 2 DIABETES

The importance of T cell cytokines, such as interferon gamma (IFNγ) and interleukin-17 in obesity is less appreciated than thoroughly examined, classical 'diabetogenic' cytokines (e.g., tumor necrosis factor-α, interleukin-6 and interleukin-1β) [22]. T helper 17 cells, the dominant source of interleukin-17, are

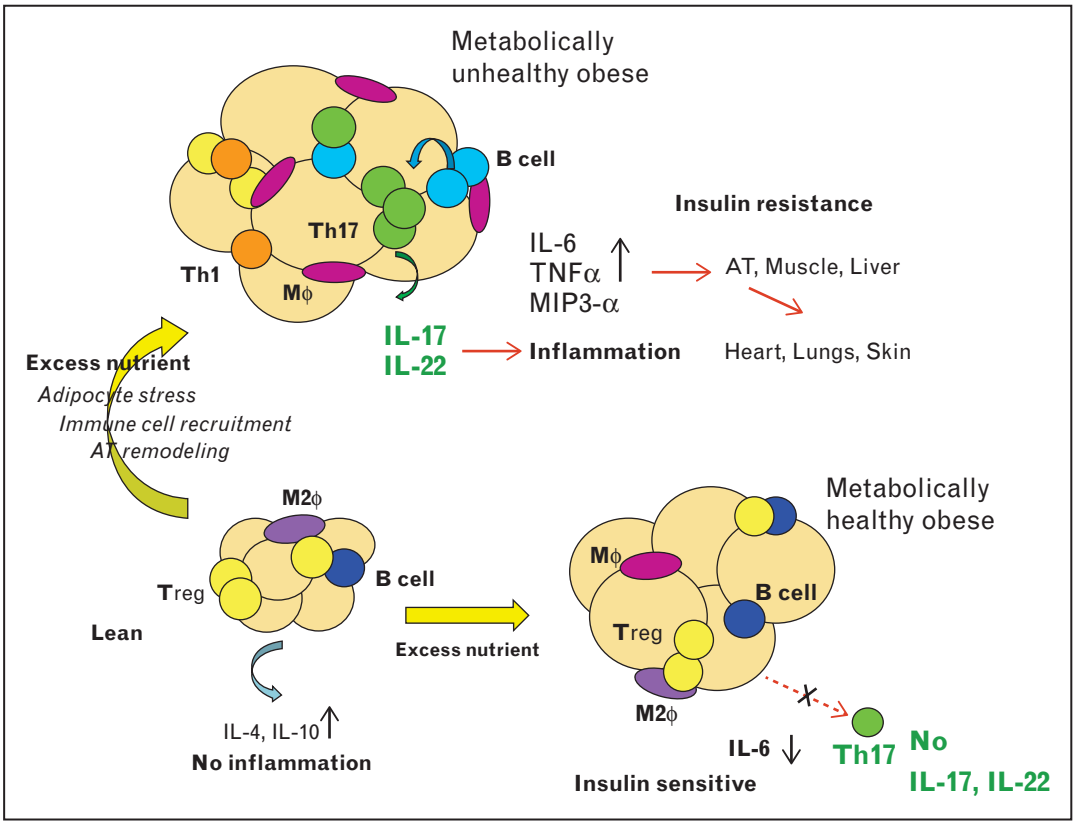

FIGURE 1. Role for immune cells in inflammatory response of metabolically healthy obese (insulin sensitive) and metabolically unhealthy (insulin resistant) obese adipose tissue. Blue = interleukin-10^{hi} B cell, light blue = interleukin-10^{lo} B cell, orange = T helper 1 cell, dark green = T helper 17 cell, yellow = Tregs, purple oval = anti-inflammatory M2 macrophages (M2ϕ), pink oval = proinflammatory M1 macrophage (Mϕ). Anti-inflammatory immune cells dominate metabolically healthy tissue, but decrease in metabolically unhealthy tissue. In contrast, proinflammatory immune cells dominate metabolically unhealthy tissue. AT, adipose tissue; IL, interleukin; MIP3-α, macrophage inflammatory protein 3 alpha; PPARγ, peroxisome proliferator-activator receptor γ; PR-DM16, positive regulatory domain 16; RANTES, regulated on activation normal T cell expressed and secreted; S100A9, soluble protein 100 α 9; Th, T helper; TNFα, tumor necrosis factor-α; Tregs, regulatory T cells.

instead recognized primarily for roles in clearance of select pathogens and for detrimental effects in autoimmune diseases, such as multiple sclerosis (mouse experimental autoimmune encephalitis) [38▪,39]. Data showing that interleukin-6 and interleukin-1β drive T helper 17 differentiation [40,41] are part of an emerging appreciation of T helper 17 expansion in obesity-associated insulin resistance/T2D. Recently, leptin, an adipokine generally increased in obesity/insulin resistance, was shown to also support T helper 17 expansion [42▪,43]. Overall, the relationship between increased interleukin-6/interleukin-1β/leptin and increased T helper 17 differentiation, coupled with the importance of T helper 17s in autoimmune diseases, may explain clinical evidence that obesity predisposes people to increased risk for inflammation-mediated autoimmune diseases, including psoriasis, rheumatoid arthritis [44], lupus [45] and multiple sclerosis [7,46].

Despite these likely mechanistic links between obesity-associated inflammation and T helper 17 cells, the T helper 17 signature cytokines interleukin-17 and interleukin-22 have been more or less dismissed in mouse models of obesity and T2D, in part because interleukin-17^{+} cells (likely T helper 17s) infiltrate murine subcutaneous rather than visceral adipose tissue in response to HFD [47], or T helper 17s dominate only after obesity/insulin resistance is established [48]. Interleukin-17 gene deletion caused an abnormal weight gain on both low fat or HFD compared with wild type controls, precluding a straight-forward interpretation of roles for T helper 17 cells in obese knockout animals [49]. This lack of strong data for interleukin-17/T helper 17 dominance in obesity-associated insulin resistance in mice has moderated excitement over possible functions of T helper 17s in human obesity/insulin resistance/T2D, although several

reports have shown interleukin-17 and interleukin-22 induced insulin resistance in chief metabolic regulators, such as hepatocytes, adipocytes and myocytes [37▪▪,49,50].

Evidence of a role for T helper 17s in human disease includes demonstrations that plasma T helper 17 cytokines and in-vitro T helper 17 activity tightly correlate with measures of glycated hemoglobin A1c levels of T2D patients [33,36,51▪]. Two recent studies also showed increased T helper 17 function (i.e., interleukin-17 and interleukin-22 production) adipose tissue-associated immune cells from well characterized obese/insulin sensitive versus obese/insulin resistant patients [37▪▪,52▪▪]. Taken together, this work suggested that some aspects of human immunometabolism should be more thoughtfully examined with animal models that more closely recapitulate human physiology. One example of such an approach is long-term (9-month) HFD feeding of genetically homogeneous C57BL6 mice, which surprisingly revealed four different metabolic response groups. These groups include the lean/insulin resistant and obese/insulin sensitive groups that are generally absent (or even culled) following standard 12–16-week feeding protocols [53]. Measuring cytokines from these metabolic groups of mice or investigation of strains of mice with resistance to diet-induced obesity [54] could facilitate progress in understanding mechanisms underlying immune cell/cytokine involvement in patients.

OBESITY, INFLAMMATION AND ASSOCIATED CORONARY ARTERY DISEASE

Western diet together with obesity, T2D, hypertension, dyslipidemia, physical inactivity and increasing average age are believed to be primary causes of CAD and CAD-associated heart failure, the leading cause of death in T2D individuals [55]. The role of inflammation in CAD has been appreciated for decades, with a relatively early appreciation that local immune cell infiltrates characterize CAD and can specifically predict risk for fatal outcome [56,57]. Immune cell infiltrates occur downstream of endothelial dysfunction, which promotes lipoprotein transcytosis from the plasma into the vessel intima. Subsequent immune cell infiltration/activation thereby links endothelial changes to cardiovascular disease/CAD [58,59]. Teasing out the complex interactions among obesity, CAD and T2D is a daunting endeavor, but the appreciation of inflammation as a critical mediator primes the field to expand the findings above thus spur fundamentally new clinical approaches.

Documented roles for macrophages, T cells and B cells in CAD suggest that obesity-associated inflammation bridges immune cell function and CAD [60–66]. Macrophages were the first immune cell recognized to play critical roles in the development of atherosclerosis, from early fatty streak formation through progression to vulnerable plaques. Mechanisms of macrophage involvement in CAD include the ability to form foam cells, and to secrete high amounts of proinflammatory, pro-CAD cytokines such as tumor necrosis factor-α, interleukin-1β, interleukin-6 and interleukin-8 [60,67–72].

Proinflammatory T cells also play critical roles in the endothelial dysfunction that precedes CAD [61–65]. IFNγ, a proinflammatory cytokine produced by both $CD4^+$ and $CD8^+$ T cells, is critical for the development of atherosclerosis. Furthermore, inhibition of T helper 1 cells, thus IFNγ and other $CD4^+$-associated cytokines, ameliorates disease in mouse models [73], independently indicating that T helper 1s promote CAD. Similarly, interleukin-17, the major cytokine produced by T helper 17s, is important for the recruitment of macrophages to developing atherosclerotic lesions [61,74,75▪], and both $CD4^+$ and $CD8^+$ T cells characterize unstable plaques (71–74). Thus, multiple lines of evidence suggest that proinflammatory T cells support obesity-associated atherosclerosis and may link CAD to T2D.

B cells are more recently appreciated mediators of CAD, although their roles are more complex than the proatherogenic functions of macrophages and T cells. B cell depletion protects against CAD in model animals [66], suggesting that B cells are pathogenic. In contrast, some studies show that B cells protect against atherosclerosis [76]. The latter interpretation is consistent with the demonstration that removal of the normal splenic reservoir of B cells renders patients more susceptible to CAD [77]. Although exact mechanisms are not known, these seemingly contradictory findings on roles for B cells in atherosclerosis may be explained by demonstrations that B cells can initially protect against inflammatory disease, but then can change their activity, promoting pathogenesis [10▪▪,34,78]. For example, the loss of B cell interleukin-10 in T2D discussed above, coupled with a T2D-associated increase in the ability of B cells to produce CAD-associated interleukin-8 [72,79], is consistent with a disease-associated gain of pathogenic B cell function. B cells also promote pericardial adipose tissue expansion and inflammation in a mouse model of obesity-associated insulin resistance [10▪▪]. Taken together, these reports suggest that B cells from obese/insulin resistant patients support CAD-associated inflammation

through multiple mechanisms, such as promoting proinflammatory cytokine production including high interleukin-8 and low interleukin-10 release [34,69,80,81], supporting adipose tissue expansion in obesity [10▪▪] and promoting proinflammatory T helper 17 cells [6,10▪▪,82]. Both the activities of individual immune cells and the cross-talk among immune cell subsets raise the clinically critical possibility that immunomodulatory drugs, such as the generally well tolerated B cell depletion drug rituximab [83,84], may have unappreciated efficacy in the prevention of obesity-associated CAD [85].

THE ROLE OF PERICARDIAL ADIPOSE TISSUE IN LOCAL INFLAMMATION AND CORONARY ARTERY DISEASE

Although adipose depots all increase in volume with obesity, fat deposits in different anatomical regions show depot-associated levels of immune cell infiltration and inflammation, and thus differentially associate with disease. To generalize, subcutaneous adipose tissue is more metabolically 'protective', whereas pericardial and other visceral depots are highly correlated with risk for obesity-associated disease, including CAD [86–88]. The recognition that pericardial adipose tissue (pAT) physiology, which includes pAT inflammation, is a critical mediator of CAD significantly departs from the outdated assumption that pAT expansion is an uninteresting epiphenomenon of both obesity-independent and obesity-associated CAD.

Numerous analyses point to the importance of pAT expansion and concomitant inflammation in obesity/insulin resistance-associated CAD. pAT from patients with CAD has increased inflammatory hallmarks compared with pAT from patients undergoing non-CAD heart procedures [72,89–91]. Notably, pAT volume also associates with systemic inflammation, as measured by interleukin-6 and C-reactive protein levels [92]. Also, CAD is more tightly associated with the amount of pAT in lean individuals than with a variety of more 'accepted' CAD risk factors, including body fat distribution [93]. pAT volume is a strong independent risk factor for CAD severity [88], and positively associates with calcified coronary plaque [94,95] which suggests that pAT may exert local toxic effects on the coronary vasculature [96]. Thus, it is unsurprising that the amount of pAT negatively correlates with cardiac output and stroke volume [97]. Additionally, epidemiological studies show that relatively high pAT volume ($>300\,cm^3$) associates with a four-fold increased risk of CAD, whereas smoking and T2D increase CAD risk 1.6-fold and three-fold, respectively [98]. Prospective studies revealed that the volume of epicardial adipose tissue, the depot that literally coats the myocardium, is predictive of obstructive CAD even before patients develop overt disease [99▪,100,101]. Finally, perhaps the most convincing human evidence linking obesity, inflammation and CAD is analysis of monozygotic twins discordant for obesity. This study found that the obese twin had a greater epicardial adipose tissue volume, and that C-reactive protein, one surrogate for systemic inflammation, was the only one of several factors measured that significantly associated with epicardial adipose tissue volume. These studies support the conclusion that inflammation and epicardial adipose tissue volume predict risk for CAD [102▪,103] and highlight the urgency of a more comprehensive analysis of pAT physiology focused

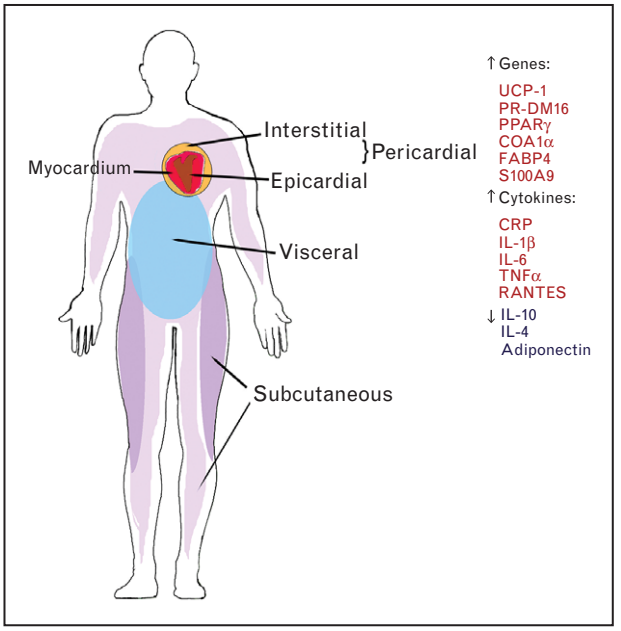

FIGURE 2. Depiction of regional adipose tissue deposition in human body and obesity-associated gene and cytokine profiles. Adipose tissue depots that preferentially expand in metabolically unhealthy (inflammatory) responses include pericardial (epicardial and interstitial) and visceral depots. Adipose tissue that preferentially expands in metabolically healthy obese individuals includes widespread subcutaneous depots. Shown are upregulated or downregulated genes, adipokines, or cytokines associated with obesity comorbidities, T2D and CAD. CAD, coronary artery disease; COA1α, cytochrome oxidase assembly 1α; CRP, C-reactive protein; FABP4, fatty acid binding protein 4; IL, interleukin; PPARγ, peroxisome proliferator-activator receptor γ; PR-DM16, positive regulatory domain 16; RANTES, regulated on activation normal T cell expressed and secreted; S100A9, soluble protein 100 α 9; TNFα, tumor necrosis factor-α; T2D, type 2 diabetes; UCP-1, uncoupling protein 1.

on inflammation to advance the long-term goal of countering CAD pathogenesis.

Together, the associations between pAT inflammation and CAD/impaired cardiovascular function, plus the known roles of obesity in adipose tissue volume and proinflammatory immune cell function [4–7,10▪▪,33,34,47,72,79,89,91,104], frame the idea that obesity-associated changes in pAT physiology link obesity and CAD and are further exacerbated in the presence of insulin resistance. However, one gap in the pAT analyses is that 'pAT' is often imprecisely defined and can denote the epicardial adipose tissue that coats the heart, the interstitial adipose tissue that coats the outside of the pericardial membrane (thus, does not directly touch the heart) or both. Both heart-proximal depots (Fig. 2) are linked to local and systemic inflammation, although each is also somewhat distinct from the other [72,89–92,105,106▪]. Epicardial adipose tissue may disproportionately impact CAD because of a shared circulation with the myocardium [107]. Regardless, a comparison of physiology of epicardial fat, interstitial fat and blood from the same individual is essential to comprehensively understand inflammation in heart-proximal adipose tissue, and will be vital for identifying pAT signatures in a more accessible tissue.

A clinically important, yet unappreciated aspect of studies on obesity, inflammation and CAD is the possibility that fundamentally different mechanisms drive CAD in lean/insulin sensitive compared to obese/insulin sensitive or obese/insulin resistant patients. This novel prospect is raised by our recent work showing mechanisms of periodontitis, another common comorbidity in obesity/insulin resistance, significantly differ between lean/insulin sensitive and obese/insulin resistant mice [108]. Lean mice developed periodontitis through a B cell-independent process, whereas periodontitis development in obese/insulin resistant mice was highly B cell-dependent. Obesity-associated inflammatory responses in periodontitis and CAD are inadequately studied in humans, although one cross-sectional study of obese subjects with and without periodontitis identified that both obesity and periodontitis increased risk for cardiovascular disease [109▪]. Future identification of correlates of CAD over a range of metabolic status with subsequent mechanistic analyses will be needed to determine whether the currently similar standard of care for CAD in lean and obese/insulin resistant patients is the best approach.

CONCLUSION

Our developing appreciation of links among obesity, inflammation and CAD will require multiple complementary approaches to leverage new concepts into translatable outcomes. Careful characterization of human patients, particularly analysis of adipose tissue distribution by measures as simple as waist-to-hip ratio, will be needed to stratify subjects that are most likely obese/metabolically healthy from those that are obese/metabolically unhealthy [93,110]. Notably, subjects with T2D are most effectively analyzed as a unique cohort, rather than being lumped into the 'obese/insulin resistant' category. Use of models that more closely parallel human disease will increase the translational significance of studies. A call for simple analysis of metabolic status in all clinical drug trials (e.g., HbA1c) would identify drugs with potential efficacy against obesity-associated CAD, although incurring minimal extra cost. Similarly, collection of (at least) serum and peripheral blood mononuclear cells from all obesity or CAD-associated clinical trials would provide samples for testing of new possibilities, such as T helper 17 dominance. Concepts developed in human sample studies, with further testing in models to refine concepts, will integrate the strengths of both bench and bedside research to allow the field to exploit our understanding of obesity-associated inflammation for clinical gains over the short term.

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Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- ▪ of special interest
- ▪▪ of outstanding interest

1. Skinner A, Skelton JA. Prevalence and trends in obesity and severe obesity among children in the United States. JAMA Pediatr 2014; 168:561–566.
2. ▪ Bastien M, Poirier P, Lemieux I, Despres JP. Overview of epidemiology and contribution of obesity to cardiovascular disease. Progress in cardiovascular diseases 2014; 56:369–381.

This review provides a clear overview of obesity contributions to cardiovascular disease with focus on abdominal adipose tissue inflammation. Authors emphasize that simplifying therapeutic objectives by directed study of higher risk obese individuals might be beneficial in cardiovascular disease prevention and relatively simple measures, such as visceral/ectopic adiposity and activity level, could identify such patients in the clinic.

3. ▪▪ Alexopoulos N, Katritsis D, Raggi P. Visceral adipose tissue as a source of inflammation and promoter of atherosclerosis. Atherosclerosis 2014; 233:104–112.

Authors have compiled and summarized research studies that link intraabdominal and epicardial adipose tissue to CAD. They recommend that noninvasive computed tomography, MRI or echocardiography could be employed to assess these new markers of CAD risk.

4. Xu H, Barnes GT, Yang Q, *et al.* Chronic inflammation in fat plays a crucial role in the development of obesity-related insulin resistance. J Clin Invest 2003; 112:1821–1830.
5. Weisberg SP, McCann D, Desai M, *et al.* Obesity is associated with macrophage accumulation in adipose tissue. J Clin Invest 2003; 112:1796–1808.
6. Winer DA, Winer S, Shen L, *et al.* B cells promote insulin resistance through modulation of T cells and production of pathogenic IgG antibodies. Nat Med 2011; 17:610–617.
7. Winer S, Paltser G, Chan Y, *et al.* Obesity predisposes to Th17 bias. Eur J Immunol 2009; 39:2629–2635.
8. Feuerer M, Herrero L, Cipolletta D, *et al.* Lean, but not obese, fat is enriched for a unique population of regulatory T cells that affect metabolic parameters. Nat Med 2009; 15:930–939.
9. Nishimura S, Manabe I, Nagasaki M, *et al.* CD8+ effector T cells contribute to macrophage recruitment and adipose tissue inflammation in obesity. Nat Med 2009; 15:914–920.
10. ■■ Defuria J, Belkina AC, Jagannathan-Bogdan M, *et al.* B cells promote inflammation in obesity and type 2 diabetes through regulation of T-cell function and an inflammatory cytokine profile. Proc Natl Acad Sci U S A 2013; 110:5133–5138.

This study revealed a critical disease-associated interplay between B and T cells, with B cells promoting insulin resistance by multiple mechanisms, such as release of proinflammatory cytokines, stimulation of adipose tissue growth/expansion, altering T cell ratios, increasing T helper 17/T helper 1 over Treg. A major strength was to apply findings from the mouse model to identify functional similarities of peripheral B and T cells from human T2D patients.

11. Duffaut C, Galitzky J, Lafontan M, Bouloumie A. Unexpected trafficking of immune cells within the adipose tissue during the onset of obesity. Biochem Biophys Res Commun 2009; 384:482–485.
12. ■ Aroor AR, DeMarco VG, Jia G, Sowers JR. Maladaptive immune and inflammatory pathways lead to cardiovascular insulin resistance. Metabolism 2013; 62:1543–1552.

This review gives brief but detailed summaries of inflammatory mediators involved in insulin resistance and their impact on cardiovascular disease.

13. Yang M, Deng J, Yang I, *et al.* IL-10-producing regulatory B10 cells ameliorate collagen-induced arthritis via suppressing Th17 cell generation. Am J Pathol 2012; 180:2375–2385.
14. Yudkin JS, Kumari M, Humphries SE, Mohamed-Ali V. Inflammation, obesity, stress and coronary heart disease: is interleukin-6 the link? Atherosclerosis 2000; 148:209–214.
15. Cheng KH, Chu CS, Lee KT, *et al.* Adipocytokines and proinflammatory mediators from abdominal and epicardial adipose tissue in patients with coronary artery disease. Int J Obes (Lond) 2008; 32:268–274.
16. Sutherland ER. Linking obesity and asthma. Ann N Y Acad Sci 2014; 1311:31–41.
17. Davidson WJ, Mackenzie-Rife KA, Witmans MB, *et al.* Obesity negatively impacts lung function in children and adolescents. Pediatr Pulmonol 2013. [Epub ahead of print]
18. Cavailles A, Brinchault-Rabin G, Dixmier A, *et al.* Comorbidities of COPD. Eur Respir Rev 2013; 22:454–475.
19. Lashinger LM, Ford NA, Hursting SD. Interacting inflammatory and growth factor signals underlie the obesity-cancer link. J Nutr 2014; 144:109–113.
20. Lumeng CN, Saltiel AR. Inflammatory links between obesity and metabolic disease. J Clin Invest 2011; 121:2111–2117.
21. ■ Xu X, Grijalva A, Skowronski A, *et al.* Obesity activates a program of lysosomal-dependent lipid metabolism in adipose tissue macrophages independently of classic activation. Cell Metab 2014; 18:816–830.

Results reported in this study define a noninflammatory lipid lipolysis role for macrophages within adipose tissue that actually has an effect on whole adipose tissue metabolism. These data point to a novel aspect of immune cell function within adipose tissue that might have an unrecognized contribution to obesity/metabolic dysfunction and may open a new avenue for treatment development.

22. Cancello R, Clement K. Is obesity an inflammatory illness? Role of low-grade inflammation and macrophage infiltration in human white adipose tissue. BJOG 2006; 113:1141–1147.
23. Zeyda M, Farmer D, Todoric J, *et al.* Human adipose tissue macrophages are of an anti-inflammatory phenotype but capable of excessive pro-inflammatory mediator production. Int J Obes (Lond) 2007; 9:1420–1428.
24. ■ Amano SU, Cohen JL, Vangala P, *et al.* Local proliferation of macrophages contributes to obesity-associated adipose tissue inflammation. Cell Metab 2014; 19:162–171.

A recent study that revealed another aspect of macrophage biology with possible implications for drug targeting of adipose tissue inflammatory response.

25. Elgazar-Carmon V, Rudich A, Hadad N, Levy R. Neutrophils transiently infiltrate intra-abdominal fat early in the course of high-fat feeding. J Lipid Res 2008; 49:1894–1903.
26. Wu D, Molofsky AB, Liang HE, *et al.* Eosinophils sustain adipose alternatively activated macrophages associated with glucose homeostasis. Science 2011; 332:243–247.
27. Liu Q, Sundar K, Mishra PK, *et al.* Helminth infection can reduce insulitis and type 1 diabetes through CD25- and IL-10-independent mechanisms. Infect Immun 2009; 77:5347–5358.
28. ■ Ferrante AW Jr. Macrophages, fat, and the emergence of immunometabolism. J Clin Invest 2013; 123:4992–4993.

This review includes a recent perspective on and implications of immune cell roles in both obese and nonobese adipose tissue.

29. Nikolajczyk BS, Jagannathan-Bogdan M, Denis GV. The outliers become a stampede as immunometabolism reaches a tipping point. Immunol Rev 2012; 249:253–275.
30. Duffaut C, Zakaroff-Girard A, Bourlier V, *et al.* Interplay between human adipocytes and T lymphocytes in obesity: CCL20 as an adipochemokine and T lymphocytes as lipogenic modulators. Arterioscler Thromb Vasc Biol 2009; 29:1608–1614.
31. Kintscher U, Hartge M, Hess K, *et al.* T-lymphocyte infiltration in visceral adipose tissue: a primary event in adipose tissue inflammation and the development of obesity-mediated insulin resistance. Arterioscler Thromb Vasc Biol 2008; 28:1304–1310.
32. Wu H, Ghosh S, Perrard XD, *et al.* T-cell accumulation and regulated on activation, normal T cell expressed and secreted upregulation in adipose tissue in obesity. Circulation 2007; 115:1029–1038.
33. Jagannathan-Bogdan M, McDonnell ME, Shin H, *et al.* Elevated proinflammatory cytokine production by a skewed T cell compartment requires monocytes and promotes inflammation in type 2 diabetes. J Immunol 2011; 186:1162–1172.
34. Jagannathan M, McDonnell M, Liang Y, *et al.* Toll-like receptors regulate B cell cytokine production in patients with diabetes. Diabetologia 2010; 53:1461–1471.
35. ■ Han JM, Patterson SJ, Speck M, *et al.* Insulin inhibits IL-10-mediated regulatory T cell function: implications for obesity. J Immunol 2014; 192:623–629.

This study is the first to identify a direct effect of insulin upon Treg interleukin-10 cytokine production and reveals additional complexity of immune response in obese adipose tissue. If confirmed in humans, such effects have implications for potential avenues of drug development.

36. Sumarac-Dumanovic M, Stevanovic D, Ljubic A, *et al.* Increased activity of interleukin-23/interleukin-17 proinflammatory axis in obese women. Int J Obes (Lond) 2009; 33:151–156.
37. ■■ Fabbrini E, Cella M, McCartney SA, *et al.* Association between specific adipose tissue CD4 + T-cell populations and insulin resistance in obese people. Gastroenterology 2013; 145:e1–3.

Results appearing in this study advance the field of immunometabolism in several ways: by stratifying obese subjects, by investigating multiple metabolic cell types and by comparing detailed metabolic profiles, authors identified a skewed T cell bias toward T helper 17 that was associated with metabolic disease rather than obesity *per se*. These findings suggest new studies should include examining the T helper 17 pathway.

38. ■ Bedoya SK, Lam B, Lau K, Larkin J 3rd. Th17 cells in immunity and autoimmunity. Clin Dev Immunol 2013; 2013:986789.

Readers interested in learning more about known roles for T helper 17 cells in human pathology will find timely summaries in this review.

39. Fukaya T, Someya K, Hibino S, *et al.* Loss of sprouty4 in T cells ameliorates experimental autoimmune encephalomyelitis in mice by negatively regulating IL-1β receptor expression. Biochem Biophys Res Commun 2014; 447:471–478.
40. Korn T, Bettelli BE, Oukka M, Kuchroo VK. IL-17 and Th17 cells. Annu Rev Immunol 2009; 27:485–517.
41. Hebel K, Rudolph M, Kosak B, Chang HD, *et al.* IL-1β and TGF-β act antagonistically in induction and differentially in propagation of human proinflammatory precursor CD4+ T cells. J Immunol 2011; 187:5627–5635.
42. ■ Yu Y, Liu Y, Shi FD, *et al.* Cutting edge: leptin-induced RORgammat expression in CD4+ T cells promotes Th17 responses in systemic lupus erythematosus. J Immunol 2013; 190:3054–3058.

Experiments of the study utilize mice and human samples and demonstrate a role for leptin in stimulating differentiation of T helper 17 cells, providing further evidence of a potential link between obesity and susceptibility to autoimmune disease.

43. Amarilyo G, Iikuni N, Shi FD, *et al.* Leptin promotes lupus T-cell autoimmunity. Clin Immunol 2013; 149:530–533.
44. Russolillo Anna IS, Peluso Rosario, *et al.* Obesity and psoriatic arthritis: from pathogenesis to clinical outcome and management. Rheumatology (Oxford) 2013; 52:62–67.
45. Rizk A, Gheita T, Nassef S, Abdallah A. The impact of obesity in systemic lupus erythematosus on disease parameters, quality of life, functional capacity and the risk of atherosclerosis. Int J Rheum Dis 2012; 15:261–267.
46. Hedström AK, Lima Bomfim I, Barcellos L, *et al.* Interaction between adolescent obesity and HLA risk genes in the etiology of multiple sclerosis. Neurology 2014; 82:865–872.
47. Winer S, Chan Y, Paltser G, *et al.* Normalization of obesity-associated insulin resistance through immunotherapy. Nat Med 2009; 15:921–929.
48. Bertola A, Ciucci T, Rousseau D, *et al.* Identification of adipose tissue dendritic cells correlated with obesity-associated insulin-resistance and inducing Th17 responses in mice and patients. Diabetes 2012; 61:2238–2247.
49. Zuniga LA, Shen WJ, Joyce-Shaikh B, *et al.* IL-17 regulates adipogenesis, glucose homeostasis, and obesity. J Immunol 2010; 185:6947–6959.

50. Tang Y, Bian Z, Zhao L, *et al.* Interleukin-17 exacerbates hepatic steatosis and inflammation in nonalcoholic fatty liver disease. Clin Exp Immunol 2011; 166:281–290.

51. ▪ Sumarac-Dumanovic M, Jeremic D, Pantovic A, *et al.* Therapeutic improvement of glucoregulation in newly diagnosed type 2 diabetes patients is associated with a reduction of IL-17 levels. Immunobiology 2013; 218:1113–1118.

This study demonstrates that fairly simple measures of serum cytokine levels along with metabolic profiles thoughtfully obtained prior to and after initiation of treatment for a metabolic disease can facilitate decisions about research focus areas.

52. ▪▪ Dalmas E, Venteclef N, Caer C, *et al.* T cell-derived IL-22 amplifies IL-1beta-driven inflammation in human adipose tissue: relevance to obesity and type 2 diabetes. Diabetes 2014; 63:1966–1977.

This study has also identified a T helper 17 pathway cytokine, interleukin-22 and interleukin-1beta to be critical mediators of inflammatory responses identified within obese diabetic adipose tissue of human patients. Such recent findings support further investigation and focus on this immune pathway in humans.

53. Burcelin Re?my CV, Dacosta Anabela, Roy-Tirell Alexandrai, Thorens Bernard. Heterogeneous metabolic adaptation of C57BL/6J mice to high-fat diet. Am J Physiol Endocrinol Metab 2002; 282:E834–E842.

54. Lee KTY, Karunakaran S, Ho MM, Clee SM. PWD/PhJ and WSB/EiJ mice are resistant to diet-induced obesity but have abnormal insulin secretion. Endocrinology 2011; 152:3005–3017.

55. Kones RRU. Prevention of cardiovascular disease: updating the immensity of the challenge and the role of risk factors. Hosp Pract 19952014; 42:92–100.

56. Kohchi K, Takebayashi S, Hiroki T, Nobuyoshi M. Significance of adventitial inflammation of the coronary artery in patients with unstable angina: results at autopsy. Circulation 1985; 71:709–716.

57. Stratford N, Britten K, Gallagher P. Inflammatory infiltrates in human coronary atherosclerosis. Atherosclerosis 1986; 59:271–276.

58. Wang Z, Nakayama T. Inflammation a link between obesity and cardiovascular disease. Mediators Inflamm 2010; 2010:535918.

59. Schafer KKS. Update on the cardiovascular risk in obesity: endocrine and paracrine role of the adipose tissue. Hell J Cardiol 2011; 52:327–336.

60. Moore KJ, Tabas I. Macrophages in the pathogenesis of atherosclerosis. Cell 2011; 145:341–355.

61. Smith E, Prasad KM, Butcher M, *et al.* Blockade of interleukin-17A results in reduced atherosclerosis in apolipoprotein E-deficient mice. Circulation 2010; 121:1746–1755.

62. Butcher MJ, Gjurich BN, Phillips T, Galkina EV. The IL-17A/IL-17RA axis plays a proatherogenic role via the regulation of aortic myeloid cell recruitment. Circ Res 2012; 110:675–687.

63. Eid RE, Rao DA, Zhou J, *et al.* Interleukin-17 and interferon-gamma are produced concomitantly by human coronary artery-infiltrating T cells and act synergistically on vascular smooth muscle cells. Circulation 2009; 119:1424–1432.

64. Zhao Z, Wu Y, Cheng M, *et al.* Activation of Th17/Th1 and Th1, but not Th17, is associated with the acute cardiac event in patients with acute coronary syndrome. Atherosclerosis 2011; 217:518–524.

65. Methe H, Brunner S, Wiegand D, *et al.* Enhanced T-helper-1 lymphocyte activation patterns in acute coronary syndromes. J Am Coll Cardiol 2005; 45:1939–1945.

66. Ait-Oufella H, Herbin O, Bouaziz JD, *et al.* B cell depletion reduces the development of atherosclerosis in mice. J Exp Med 2010; 207:1579–1587.

67. Gerrity RG. The role of the monocyte in atherogenesis: II. Migration of foam cells from atherosclerotic lesions. Am J Pathol 1981; 103:191–200.

68. Gerrity RG. The role of the monocyte in atherogenesis: I. Transition of blood-borne monocytes into foam cells in fatty lesions. Am J Pathol 1981; 103:181–190.

69. Kanda T, Hirao Y, Oshima S, *et al.* Interleukin-8 as a sensitive marker of unstable coronary artery disease. Am J Cardiol 1996; 77:304–307.

70. Lachman LB, Page SO, Metzgar RS. Purification of human interleukin 1. J Supramol Struct 1980; 13:457–466.

71. Matthews N. Tumour-necrosis factor from the rabbit. II. Production by monocytes. Br J Cancer 1978; 38:310–315.

72. Sacks HS, Fain JN, Cheema P, *et al.* Depot-specific overexpression of proinflammatory, redox, endothelial cell, and angiogenic genes in epicardial fat adjacent to severe stable coronary atherosclerosis. Metab Syndr Relat Disord 2011; 9:433–439.

73. Hansson GK. Inflammation atherosclerosis, and coronary artery disease. N Engl J Med 2005; 352:1685–1695.

74. Taleb S, Herbin O, Ait-Oufella H, *et al.* Defective leptin/leptin receptor signaling improves regulatory T cell immune response and protects mice from atherosclerosis. Arterioscler Thromb Vasc Biol 2007; 27:2691–2698.

75. ▪ Chalubinski M, Wojdan K, Dorantowicz R, *et al.* Comprehensive insight into immune regulatory mechanisms and vascular wall determinants of atherogenesis - emerging perspectives of immunomodulation. Arch Med Sci 2013; 9:159–165.

This review covers work describing how additional cellular players, including T helper 17 and Treg, in the etiology of cardiovascular disease need to be incorporated into current thinking about therapeutics.

76. Caligiuri G, Nicoletti A, Poirier B, Hansson GK. Protective immunity against atherosclerosis carried by B cells of hypercholesterolemic mice. J Clin Invest 2002; 109:745–753.

77. Witztum JL. Splenic immunity and atherosclerosis: a glimpse into a novel paradigm? J Clin Invest 2002; 109:721–724.

78. Matsushita T, Yanaba K, Bouaziz JD. Regulatory B cells inhibit EAE initiation in mice while other B cells promote disease progression. J Clin Invest 2008; 118:3420–3430.

79. Sacks HS, Fain JN, Cheema P, *et al.* Inflammatory genes in epicardial fat contiguous with coronary atherosclerosis in the metabolic syndrome and type 2 diabetes: changes associated with pioglitazone. Diabetes Care 2011; 34:730–733.

80. Jagannathan M, Hasturk H, Liang Y, *et al.* TLR cross-talk specifically regulates cytokine production by B cells from chronic inflammatory disease patients. J Immunol 2009; 183:7461–7470.

81. Noronha AM, Liang Y, Hetzel JT, *et al.* Hyperactivated B cells in human inflammatory bowel disease. J Leukoc Biol 2009; 86:1007–1016.

82. Van de Veerdonk FL, Lauwerys B, Marijnissen RJ, *et al.* The anti-CD20 antibody rituximab reduces the Th17 cell response. Arthritis Rheum 2011; 63:1507–1516.

83. Salliot C, Dougados M, Gossec L. Risk of serious infections during rituximab, abatacept and anakinra treatments for rheumatoid arthritis: meta-analyses of randomised placebo-controlled trials. Ann Rheum Dis 2009; 68:25–32.

84. Linos E. Balancing the risks and benefits of rituximab. JAMA Intern Med 2013; 173:920–937.

85. Pescovitz MD, Greenbaum CJ, Krause-Steinrauf H, *et al.* Rituximab, B-lymphocyte depletion, and preservation of beta-cell function. N Engl J Med 2009; 361:2143–2152.

86. Payne GA, Kohr MC, Tune JD. Epicardial perivascular adipose tissue as a therapeutic target in obesity-related coronary artery disease. Br J Pharmacol 2012; 165:659–669.

87. Konishi M, Sugiyama S, Sugamura K, *et al.* Association of pericardial fat accumulation rather than abdominal obesity with coronary atherosclerotic plaque formation in patients with suspected coronary artery disease. Atherosclerosis 2010; 209:573–578.

88. Taguchi R, Takasu J, Itani Y, *et al.* Pericardial fat accumulation in men as a risk factor for coronary artery disease. Atherosclerosis 2001; 157:203–209.

89. Mazurek T, Zhang L, Zalewski A, *et al.* Human epicardial adipose tissue is a source of inflammatory mediators. Circulation 2003; 108:2460–2466.

90. Iacobellis G, Pistilli D, Gucciardo M, *et al.* Adiponectin expression in human epicardial adipose tissue in vivo is lower in patients with coronary artery disease. Cytokine 2005; 29:251–255.

91. Fain JN, Sacks HS, Bahouth SW, *et al.* Human epicardial adipokine messenger RNAs: comparisons of their expression in substernal, subcutaneous, and omental fat. Metabolism 2010; 59:1379–1386.

92. Tadros TM, Massaro JM, Rosito GA, *et al.* Pericardial fat volume correlates with inflammatory markers: the Framingham Heart Study. Obesity (Silver Spring) 2010; 18:1039–1045.

93. Denis GV, Hamilton JA. Healthy obese persons: how can they be identified and do metabolic profiles stratify risk? Curr Opin in Endocrinol Diabetes Obes 2013; 20:369–376.

94. Ding J, Kritchevsky SB, Harris TB, *et al.* The association of pericardial fat with calcified coronary plaque. Obesity (Silver Spring) 2008; 16:1914–1919.

95. McClain J, Hsu F, Brown E, *et al.* Pericardial adipose tissue and coronary artery calcification in the Multiethnic Study of Atherosclerosis (MESA). Obesity (Silver Spring) 2013; 21:1056–1063.

96. Rosito GA, Massaro JM, Hoffmann U, *et al.* Pericardial fat, visceral abdominal fat, cardiovascular disease risk factors, and vascular calcification in a community-based sample: the Framingham Heart Study. Circulation 2008; 117:605–613.

97. Ruberg FL, Chen Z, Hua N, *et al.* The relationship of ectopic lipid accumulation to cardiac and vascular function in obesity and metabolic syndrome. Obesity (Silver Spring) 2010; 18:1116–1121.

98. Greif M, Becker A, von Ziegler F, *et al.* Pericardial adipose tissue determined by dual source CT is a risk factor for coronary atherosclerosis. Arterioscler Thromb Vasc Biol 2009; 29:781–786.

99. ▪ Neeland IJ, Gupta S, Ayers CR, *et al.* Relation of regional fat distribution to left ventricular structure and function. Circ Cardiovasc Imaging 2013; 77:1835–1844.

Authors report recent findings of fat distribution assessed by both MRI and dual energy X-ray absorptiometry in a cross-sectional analysis of more than 2700 participants in the Dallas Heart Study. Their attention to regional adipose tissue differences and detailed morphometry revealed that within obese subjects, significant association between visceral adipose tissue and concentric remodeling of the heart was observed, although subcutaneous lower body adipose tissue was protective for left ventricular function, no such association between hemodynamics and subcutaneous abdominal adipose tissue was found.

100. Divers J, Wagenknecht LE, Bowden DW, *et al.* Regional adipose tissue associations with calcified atherosclerotic plaque: African American-diabetes heart study. Obesity (Silver Spring) 2010; 18:2004–2009.
101. Imai A, Komatsu S, Ohara T, *et al.* Visceral abdominal fat accumulation predicts the progression of noncalcified coronary plaque. Atherosclerosis 2012; 222:524–529.
102. ■ Liang Kae-Woei TI-C, Lee Wen-Jane, Lee I-Te, *et al.* MRI measured epicardial adipose tissue thickness at the right AV groove differentiates inflammatory status in obese men with metabolic syndrome. Obesity (Silver Spring) 2012; 20:525–532.
This study reports how MRI may be utilized to assess cardiovascular risk in patients by specific measurement of the epicardial adipose tissue, right atrioventricular groove.
103. Granér M, Seppälä-Lindroos A, Rissanen Aila, *et al.* Epicardial fat, cardiac dimensions, and low-grade inflammation in young adult monozygotic twins discordant for obesity. Am J Cardial 2012; 109:1295–1302.
104. Wassel CL, Laughlin GA, Araneta MR, *et al.* Associations of pericardial and intrathoracic fat with coronary calcium presence and progression in a multiethnic study. Obesity (Silver Spring) 2013; 21:1704–1712.
105. Iacobellis G. Epicardial and pericardial fat: close, but very different. Obesity (Silver Spring) 2009; 17:625.
106. ■ Sacks HS, Fain JN, Bahouth SW, *et al.* Adult epicardial fat exhibits beige features. J Clin Endocrinol Metab 2013; 98:E1448–E1455.
In the interest of defining additional brown fat-like therapeutic targets, this study determined gene expression profiles of heart-associated fat depots. In particular, epicardial adipose tissue had relatively higher UCP-1 gene transcripts compared with visceral pericardial fat, consistent with a 'beige' fat cell lineage.
107. Sacks HS, Fain JN. Human epicardial adipose tissue: a review. Am Heart J 2007; 153:907–917.
108. Zhu M, Belkina AC, Defuria J, *et al.* B cells promote obesity-associated periodontis and oral pathology-associated inflammation. J Leukoc Biol 2014. [Epub ahead of print]
109. ■ Pires JR, Dos Santos IP, de Camargo LF, *et al.* Framingham cardiovascular risk in patients with obesity and periodontitis. J Indian Soc Periodontol 2014; 18:14–18.
One of only a few recent human studies in which metabolic and periodontitis parameters were evaluated for cardiac risk in obese subjects. Using the Framingham Score, both obesity and periodontitis positively correlated with increased risk for cardiovascular disease.
110. Rossi R, Iaccarino D, Nuzzo A, *et al.* Influence of body mass index on extent of coronary atherosclerosis and cardiac events in a cohort of patients at risk of coronary artery disease. Nutr Metab Cardiovasc Dis 2011; 21:86–93.