Inflammation, diabesity and cardiovascular disease: Clinical relevance

Rhodri King, Ramzi Ajjan

Cardiovascular disease is a major cause of death in people with type 2 diabetes, due to higher levels of cardiovascular risk factors such as hypertension and dyslipidaemia, as well as an increased propensity for atherosclerotic plaque formation and a pro-thrombotic environment. Inflammation has been increasingly recognised as a common feature of obesity, type 2 diabetes and cardiovascular disease and it is thought that a reduction of inflammation may be a novel therapeutic approach in this population. This article outlines the role of inflammation within diabesity and cardiovascular disease, its clinical implications and possible therapies.

The rising prevalence of obesity worldwide is inevitably leading to an increase in type 2 diabetes, characterised by an insulin-resistant state. Type 2 diabetes and obesity greatly impact on morbidity and mortality, with increased incidence of cancers, cardiovascular disease (CVD), hypertension, respiratory infections and sleep apnoea. As such, the rise of diabesity has major health and economic implications.

Cardiovascular disease remains a major cause of death in this population and its development is related to clustering of cardiovascular risk factors (hypertension, dyslipidaemia) along with an increased propensity for atherosclerotic plaque formation and a pro-thrombotic environment (Hess and Grant, 2011).

Inflammation is increasingly recognised as a common feature of obesity, type 2 diabetes and CVD, which has led to the belief that a reduction of inflammation may be a novel therapeutic approach. This review will outline the role of inflammation in diabesity and CVD, its clinical implications and possible therapies.

A brief history of inflammation in diabesity

The association between inflammation and diabetes was first noted around the turn of the last century, with evidence that sodium salicylate lowered levels of glycosuria in people with "mild" forms of diabetes. This concept failed to fully capture the imagination of the scientific world at the time, and it was not until the end of the 1950s that further evidence emerged. High-dose aspirin was found to improve blood glucose control in people with type 2 diabetes, such that insulin treatment could be withdrawn (Shoelson et al, 2006). However, researchers attempted to link their observations to the secretion of insulin rather than resistance to its action and therefore the association between inflammation and diabetes was once again dismissed.

The discovery in the 1990s that adipose cells were capable of secreting the pro-inflammatory cytokine, tumour necrosis factor-alpha (TNF-alpha), which was subsequently able to induce insulin resistance, revolutionised thinking at the time (Hotamisligil et al, 1993). There was now direct evidence that adipose cells were metabolically active, rather than mere energy storage cells with the potential to impair cellular responses to insulin and therefore ultimately lead to the hyperinsulinaemic hyperglycaemic state characteristic of type 2 diabetes.

Numerous other cytokines (termed "adipokines") and proteins are now known to be secreted by

Citation: King R, Ajjan R (2013) Inflammation, diabesity and cardiovascular disease: Clinical relevance. *Diabesity in Practice* **2**: 29–35

Article points

1. Inflammation has been increasingly recognised as a common feature of obesity, type 2 diabetes and cardiovascular disease. Reduction of inflammation may be a novel therapeutic approach for these conditions.
2. Some cytokines and proteins secreted by adipose tissue can modulate the inflammatory or thrombotic environment. Many of these cytokines are elevated in diabesity and cardiovascular disease.
3. Diabesity is associated with changes in several proteins involved in coagulation and fibrinolysis.
4. Newer therapies have been trialled, including agents that inhibit parts of the complement pathway, anti-TNF-alpha antibody medications and salicylates.

Key words

- Cardiovascular disease
- Inflammation

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Page points

1. Adipose cells secrete cytokines and proteins which can modulate the inflammatory/thrombotic environment. The majority of these cytokines are elevated in diabesity and cardiovascular disease.
2. Elevated levels of pro-inflammatory cytokines exert varied effects, including increased levels of monocytes and macrophages. This is associated with insulin resistance.
3. Cytokines promote atherogenesis and the pro-inflammatory adipokines associated with diabesity enhance atherosclerosis.
4. Diabesity is associated with changes in several proteins involved in coagulation and fibrinolysis.

adipose tissue, which can modulate the inflammatory or thrombotic environment, including interleukin 6 (IL-6), C-reactive protein (CRP), leptin, resistin, adiponectin, plasminogen activator inhibitor-1 (PAI-1), monocyte chemoattractant protein-1 (MCP-1), complement protein C3 and many others (Galic et al, 2010).

The majority of these cytokines are elevated in diabesity and CVD and proteins, such as CRP, IL-6 and C3, have been associated with the development of type 2 diabetes and vascular complications (Thorand et al, 2003; Ajjan et al, 2005; Engstrom et al, 2005).

Current theories linking diabesity and inflammation revolve around the nuclear factor NF-kappa B, which, once activated by I kappa B kinase (IKK), promotes and maintains inflammation; this pathway is stimulated by obesity (Goldfine et al, 2011). Elevated levels of circulating pro-inflammatory cytokines exert varied effects within different tissue cells. Within hepatocytes and adipocytes, there is increased monocyte recruitment and macrophage formation leading to further cytokine secretion. This is associated with insulin resistance through impairment of adipocytes differentiation and insulin signalling along with hepatic and skeletal muscle lipid accumulation (Goossens, 2008). *Figure 1* illustrates this process.

Cytokines promote atherogenesis within endothelial cells (ECs). The process of atherosclerotic plaque formation is in itself an inflammatory process and so it is unsurprising that the pro-inflammatory adipokines associated with diabesity enhance atherosclerosis. EC dysfunction is the first stage of plaque formation and is accelerated by circulating cytokines. Certain adhesion molecules are up-regulated within the EC, attracting cells such as monocytes which subsequently oxidise LDL particles, eventually forming foam cells and then fatty streaks. There is a reduction in the synthesis of nitric oxide (NO), a key regulator of vascular tone, which has further deleterious effects on EC function. The production of lipid-rich plaques also

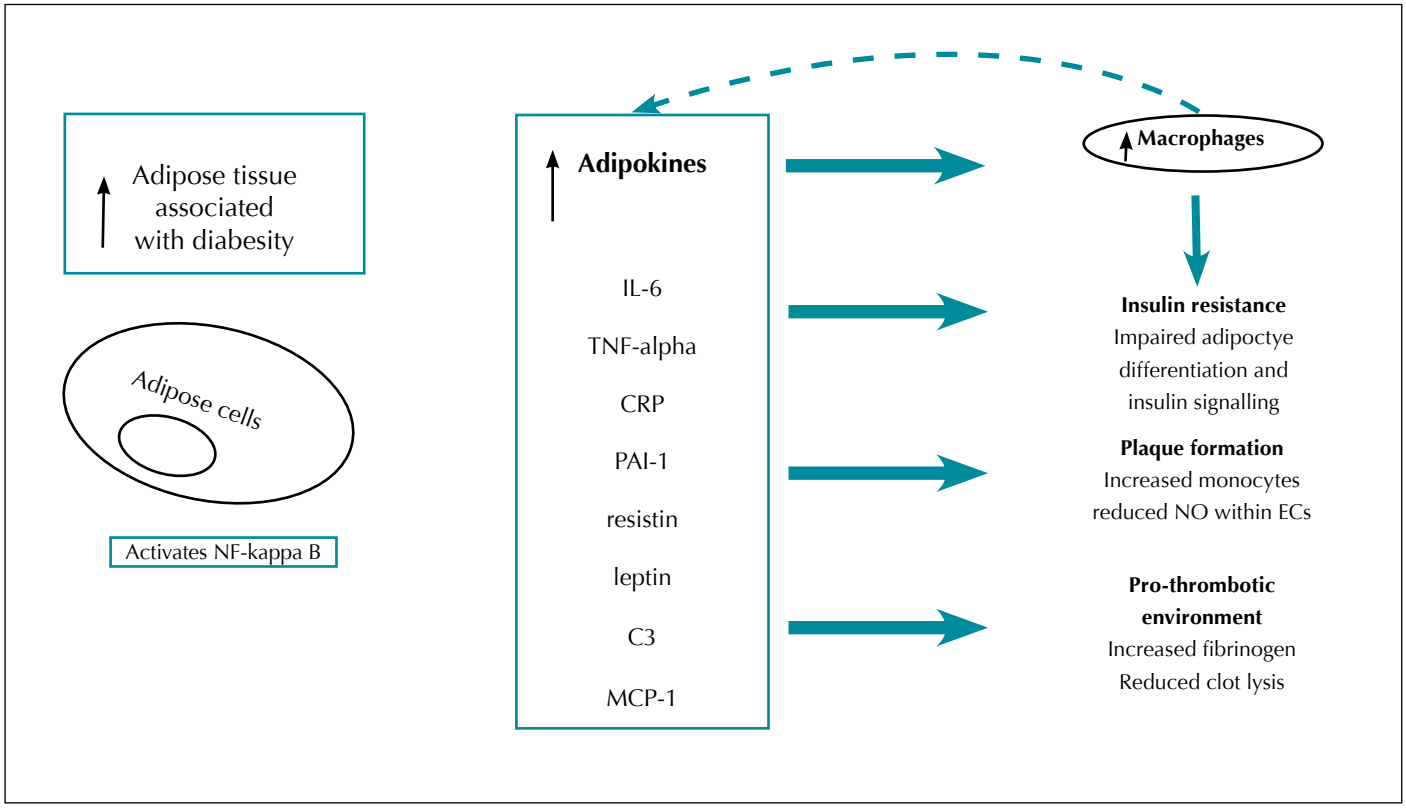

Figure 1. Excess adipose tissue activates NF- kappa B causing increased expression of adipokines. There is increased plaque formation within ECs and a pro-thrombotic environment through increased hepatic fibrinogen production and reduced clot lysis. CRP=C-reactive protein; EC=endothelial cells; IL-6=interleukin 6; MCP-1=monocyte chemoattractant protein-1; NO=nitric oxide; PAI-1=plasminogen activator inhibitor-1; TNF-alpha=tumour necrosis factor-alpha.

makes them more vulnerable to rupture leading to myocardial infarction or acute cerebrovascular events. It is also likely that the complement pathway is activated during myocardial ischaemia as numerous components of the complement pathway, such as C3, C5a and the membrane attack complex, along with CRP and macrophages, have been identified within atheromatous plaques of people with unstable angina compared with stable disease (Kostner et al, 2006; Meuwissen et al, 2006).

Diabesity associated with a pro-thrombotic environment

Diabesity is associated with quantitative and qualitative changes in several proteins involved in coagulation and fibrinolysis (*Table 1*). Elevated levels of fibrinogen and tissue factor (TF) are observed, which leads to a pro-thrombotic state. This is compounded by elevated levels of the fibrinolytic inhibitor PAI-1 resulting in prolonged lysis of clots that are formed following atherosclerotic plaque rupture. Adipokines are again implicated in this imbalance in haemostatic proteins through inducing hepatic production of both fibrinogen and PAI-1.

Increased levels of fibrinogen result in the formation of blood clots that are more tightly packed with fewer pores in between fibres, which makes the clot less susceptible to fibrinolysis (*Figure 2*). Fibrinogen also undergoes glycation as a result of hyperglycaemia, which produces similar lysis-resistant clots (Dunn et al, 2005; Pieters et al, 2007).

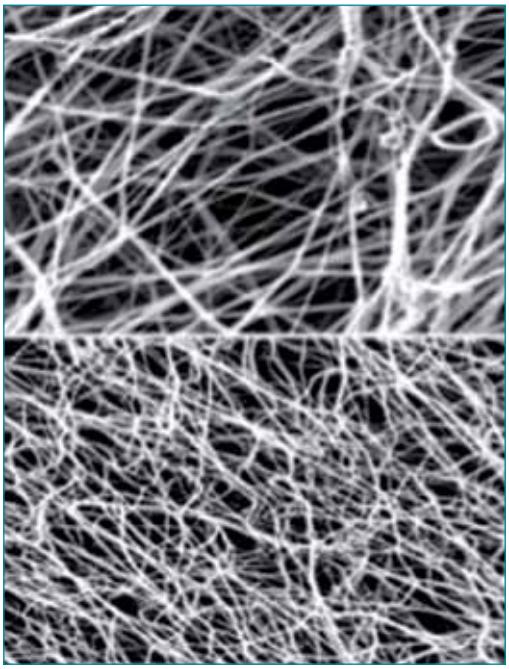

Figure 2. Electron microscopy of fibrin clots. Top image: healthy control; bottom image: person with diabetes. Diabetes clot is more dense with fewer pores between fibres. King and Ajjan (unpublished).

Inflammation and the coagulation pathway are closely linked via proteins from the complement pathway. Several complement proteins, such as C1q, C3, C4 and C5b-9, have been identified within fibrin clots (Niculescu and Rus, 1999), and the increased incorporation of C3 that occurs in people with diabetes produces a prolongation of clot lysis *in vitro* (Hess et al, 2012; Howes et al, 2012).

"Diabesity is associated with quantitative and qualitative changes in several proteins involved in coagulation and fibrinolysis"

Table 1. Changes in haemostatic proteins in diabetes.

Haemostatic protein	Function	Changes seen with diabetes	Effect of changes
Thrombin	Converts fibrinogen to fibrin	Increased levels	Altered clot structure
Fibrinogen	Forms fibrin clot	Increased levels	Altered clot structure
		Increased glycation	Decreased fibrinolysis
TF	Initiates clotting cascade	Increased levels	Increased thrombosis
PAI-1	Inhibits production of plasmin	Increased levels	Decreased fibrinolysis
FVII	Forms complex with TF	Increased levels	Increased thrombosis
FVIII and vWf complex	Binds platelets	Increased levels	Increased platelet activation

FVII=factor VII; FVIII=factor VIII; PAI-1=plasminogen activator inhibitor-1; TF=tissue factor; vWf=von Willebrand factor.

Page points

1. Numerous studies have demonstrated a reduction in inflammatory markers following improvement of diabetes control.
2. Research into the use of TNF-alpha to improve insulin sensitivity and glucose homeostatis is, so far, inconclusive in humans.
3. Salicylates have been shown to reduce glucose in obese insulin-resistant mice. However, there is a risk of gastrointestinal bleeding with long-term use.

Clinical implications

Although there remains some questions regarding the mechanisms surrounding inflammation, diabesity and CVD, there is little doubt that they are linked. Numerous studies have demonstrated a reduction in inflammatory markers following improvement of diabetes control using oral hypoglycaemic agents, and following weight loss through lifestyle modifications or bariatric surgery (Shoelson et al, 2007). But what about targeting the underlying inflammation as a potential for new therapeutic agents to improve glycaemic control and reduce cardiovascular risk and mortality? The use of systemic glucocorticoids, such as prednisolone, is well known to have the opposite effect in people with diabetes, in that it causes hyperglycaemia, and other non-steroidal anti-inflammatory agents also have no hypoglycaemic effect. These observations may be explained by the different cellular mechanisms causing the chronic, indolent inflammation of diabesity and the acute inflammatory response seen in acute infections and autoimmune conditions, for example.

Newer therapies

Complement inhibition

Agents that inhibit certain components of the complement pathway have been trialled as a treatment for coronary artery disease. Trials using pexelizumab, an anti-C5 monoclonal antibody, to block the action of C5 following myocardial infarction and post coronary artery bypass grafting have had mixed results with inconsistent reductions in mortality and major cardiac events (Granger et al, 2003; Verrier et al, 2004; Armstrong et al, 2007). The use of a C1 esterase inhibitor, TP10, has also been trialled. Given as a bolus at least 6 hours following the onset of acute myocardial infarction symptoms and subsequent infusion for 48 hours, it demonstrated reductions in the release of myocardial enzymes troponin T and creatine kinase MB, elevated levels of C1 inhibitor activity and reduced levels of C4 (Diris et al, 2002). The mixed results of these trials in the acute setting may indicate that inhibition of complement may be better directed at preventing the progression of unstable atherosclerotic plaques (Speidl et al, 2011).

TNF-blockade

Anti-TNF-alpha antibody medications, such as etanercept and infliximab, have been used successfully in inflammatory conditions, such as rheumatoid arthritis, for several years. Given the association between diabesity and elevated TNF-alpha, it may be deduced that these agents could improve insulin sensitivity and glucose homeostasis.

Animal studies have yielded positive results, which, to date, have not been translated into human studies. Markers of inflammation are reduced but results regarding glucose control have been conflicting in obese individuals with type 2 diabetes or insulin resistance, and may be related to variations in duration and dose of treatment, and in modes of administrating the medication (oral versus intravenous) seen in the different studies (Ofei et al, 1996; Paquot et al, 2000; Dominguez et al, 2005; Gonzalez-Gay et al, 2006). It may also be that many other mechanisms, over and above TNF-alpha, are involved in the inflammation associated with diabesity. A recent study using higher doses of oral etanercept for a longer duration demonstrated lower levels of fasting glucose in obese subjects with previous abnormal glucose control (Stanley et al, 2011). Further studies may be warranted to determine if these agents have potential as treatment options in diabesity.

Salicylates

As discussed previously, aspirin at high doses and sodium salicylate have demonstrated glucose-lowering capabilities, which has been confirmed in obese, insulin-resistant mice. However, the risk of serious gastrointestinal bleeding associated with long-term use of high-dose aspirin makes it an unsuitable therapy for people with diabetes. Other non-acetylated salicylates, such as sodium salicylate and the prodrug salsalate, have a much safer profile and work through inhibition of NF-kappa B (Goldfine et al, 2011).

Small pilot studies using salsalate (used in the US for joint pain) in obese, insulin-resistant people and those with type 2 diabetes demonstrated several favourable findings, including reduced glucose and triglyceride levels and elevated levels of adiponectin (cardioprotective cytokine), along with increased insulin secretion. These results led to a larger, multi-centre randomised clinical trial of three doses of salsalate versus placebo in people with type 2 diabetes conducted over 14 weeks. The three different doses each reduced levels of HbA_{1c}, fasting blood glucose and triglyceride levels, and increased adiponectin levels. Adverse events from both the pilot and randomised

trials included tinnitus at higher doses and minor elevation in urinary albumin secretion (Goldfine et al, 2010; Goldfine et al, 2011). A trial of salsalate with more people over a longer period of time is under way to determine whether it has a place in the treatment of type 2 diabetes and results of the primary outcome measure (change in HbA_{1c} from baseline to 48 weeks) should be available soon. A further study by the same group is looking at the effect of salsalate on EC function and coronary artery plaques using computed tomography angiography (Goldfine et al, 2011).

Conclusion

Treatment strategies available for diabesity are aimed at weight loss, through diet and lifestyle modification or bariatric surgery, and reducing hyperglycaemia by increasing insulin secretion, improving insulin resistance or through exogenous insulin. The ultimate goal of treatment is to reduce the risks of microvascular and macrovascular complications, and avoid hypoglycaemia. The newer insulin secretagogues, glucagon-like peptide-1 (GLP-1) agonists and dipeptidyl peptidase 4 (DPP-4) inhibitors, have been successful since their introduction and demonstrate the importance of exploring alternative pathways to achieve our goal. A better understanding of the role of inflammation in diabetes and obesity will hopefully provide novel therapeutic options and another addition to a physician's armamentarium. ■

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"An increased understanding of the role of inflammation in diabetes and obesity, will hopefully provide novel therapeutic options."