Could insulin protect against atherosclerosis?

Sharon Dorrell, Freelance writer

Scientists in the USA have discovered that insulin has acute anti-inflammatory properties and could inhibit the development of atherosclerosis (Box 1)[1]. In a recent study, Paresh Dandona (State University of New York at Buffalo, NY, USA) and colleagues demonstrated that insulin both inhibits the production of inflammatory factors and adhesion molecules involved in atherosclerosis and stimulates the production of anti-inflammatory factors.

They administered four-hour infusions of insulin plus dextrose to ten obese, insulin-resistant patients, taking care to maintain blood glucose at baseline levels throughout the infusion. Blood samples were collected before the infusion and at two, four and six hours during the experiment, and the levels of several factors associated with inflammation and the development of atherosclerosis were measured.

Anti-inflammatory insulin

Nuclear factor-κB (NFκB) is the key cellular signal that induces transcription of the inflammatory cytokines, adhesion molecules and enzymes that generate damaging reactive oxygen intermediates (ROIs). Dandona and colleagues found that levels of NFκB in mononuclear cells (MNCs), which carry insulin receptors, had decreased at two hours and continued to decrease during the four-hour infusion, but returned to baseline levels at six hours, two hours after the infusion was stopped.

NFκB activity is inhibited by the protein IκB, which binds to NFκB in the cytosol and prevents it from reaching the nucleus. IκB levels increased during the experiment and remained high even after six hours. Such changes, say the researchers, are characteristic of anti-inflammatory effects at both the cellular and molecular level. As expected, MNC production of ROIs decreased in parallel with the reduction in NFκB activity during the insulin infusion and had begun to return to baseline levels by six hours.

NADPH oxidase is an enzyme that produces the superoxide radical from molecular oxygen. Significantly, levels of $p47^{phox}$ subunit, the key protein of the NADPH oxidase complex, also decreased during the insulin infusion, but reverted to baseline levels at six hours. According to the researchers, ROIs (including the superoxide radical) stimulate NFκB-mediated inflammation and suppression of the NADPH complex by insulin contributes to the reduction in NFκB levels[1]. They add that levels of the $p47^{phox}$ subunit also decrease during administration of the anti-inflammatory drug hydrocortisone[1].

NFκB also stimulates the generation of ROIs and, therefore, the reduction in activity of this transcription factor will also lead to a decrease in the level of ROIs and thus a reduction in inflammation. Indeed, the blood levels of other factors closely associated with inflammation and atherosclerosis – soluble intercellular adhesion molecule-1 (sICAM-1), monocyte chemoattractant protein-1 (MCP-1) and plasminogen activator inhibitor-1 (PAI-1) – were also decreased during insulin infusion.

The group administered dextrose-only and saline-only control infusions but found only a small increase in the generation of ROIs during the dextrose infusion. No

Box 1. Development of atherosclerosis

Atherosclerosis is an inflammatory process triggered by an interaction between dysfunctional vascular endothelial cells and monocytes[a,b]. Endothelial dysfunction might be caused by mechanical injury or biochemical and immunological imbalances in the endothelial cells.

When endothelial cells are damaged, mononuclear cells (MNCs) and monocytes attach to the damaged cells and are closely involved in inflammation of the blood vessel walls. According to Dandona and colleagues, the generation of reactive oxygen intermediates by these cells leads to further cellular damage, lipid peroxidation, protein oxidation and damage to DNA[c]. The activation of monocytes and lipid peroxidation, in particular, are processes also known to lead to foam-cell formation, a key step in the development of fatty streaks and atherosclerosis. MNCs and monocytes, say the researchers, are also known to be active in diabetes.

References

a Ross, R. (1999) Atherosclerosis – an inflammatory disease. *New Engl. J. Med.* 340, 115–126
b Schwartz, C.J. *et al*. (1985) Atherosclerosis as an inflammatory process. The roles of the monocyte-macrophage. *Ann. New York Acad. Sci.* 454, 115–120
c Dandona, P. *et al*. (2001) Insulin inhibits intranuclear factor κB and stimulates IκB in mononuclear cells in obese subjects: evidence for an anti-inflammatory effect? *J. Clin. Endocrinol.* 86, 3257–3265

1359-6446/01/$ – see front matter PII: S1359-6446(01)02011-6

other changes were observed, but this small change is consistent with the anti-inflammatory effects demonstrated during insulin infusion.

Glitazones

In a previous study, Dandona and colleagues demonstrated that troglitazone, one of the thiazolidinedione insulin sensitizer agents (also called the glitazones), reduced inflammatory activity in obese people[2]. They found that these changes were associated with vascular improvements consistent with those observed during long-term insulin therapy[3]. 'Thus, insulin and the insulin sensitizers exert an anti-inflammatory effect that, in the long term, may prove to be anti-atherogenic,' the researchers conclude.

Future clinical studies

Dandona and colleagues suggest future investigations into the inflammatory effects of insulin infusion therapy in patients with acute myocardial infarction. Such patients are known to benefit from insulin, say the researchers, possibly because insulin inhibits lipolysis and the production of prothrombotic free fatty acids. However, if free fatty acids also possess inflammatory activity, insulin might have another mechanism of action. Furthermore, this study also suggests that insulin therapy might play a greater role in the treatment of patients with type 2 diabetes, which is associated with a high risk of atherosclerosis and its complications.

References

1 Dandona, P. *et al.* (2001) Insulin inhibits intranuclear factor κB and stimulates IκB in mononuclear cells in obese subjects: evidence for an anti-inflammatory effect? *J. Clin. Endocrinol.* 86, 3257–3265

2 Garg, R. *et al.* (2000) Troglitazone reduces reactive oxygen species generation by leukocytes and lipid peroxidation and improves flow-mediated vasodilatation in obese subjects. *Hypertension* 36, 430–435

3 Vehkavaara, S. *et al.* (2000) Insulin therapy improves endothelial function in type 2 diabetes. *Arterioscler. Thromb. Vasc. Biol.* 20, 545–550

New antibody therapies for HIV

Martina Habeck, Freelance writer

Novel monoclonal antibody-based therapies for the treatment of HIV are currently being evaluated in the laboratory and, increasingly, in clinical trials. Tanox (Houston, TX, USA) has recently announced a Phase I clinical trial of a monoclonal antibody that works by binding to the CD4 receptor on the host-cell surface. Meanwhile, a team of researchers at Rockefeller University (New York, NY, USA) has just published new research[1] that could lead to an alternative antibody-based therapy that would prevent or reverse mechanisms of HIV pathogenicity attributed to the HIV Tat protein, a protein that would otherwise destroy CD4 cells and lead to the progression of HIV to AIDS.

Preventing viral entry

HIV-1 entry into the host cell requires the binding of glycoprotein 120 (gp120) to the CD4 receptor on the host-cell surface. This interaction triggers a conformational change in gp120 that exposes its co-receptor binding site. Once gp120 has attached itself to both the CD4 receptor and its co-receptor, membrane fusion becomes possible and the virus can enter the host cell.

The monoclonal antibody TNX-355 (formerly Hu5A8) interferes with this process by binding to the extracellular domain 2 of the CD4 receptor. This does not inhibit the initial attachment of the virus to the CD4 protein, because the virus binds to domain 1. However, it blocks the conformational change of gp120, thereby preventing viral entry into the host cell.

To assess whether a humanized form of the antibody would be an effective and well-tolerated treatment for HIV, researchers at Tanox and elsewhere have done extensive preclinical tests[2]. William Shanahan, Chief Medical Officer at Tanox says that their studies showed that the antibody is effective *in vitro* at blocking infection across a broad spectrum of isolates of all the major HIV-1 subtypes. Further studies suggest that TNX-355 is unlikely to be immunogenic in humans because it has a fully human IgG core and the Fab (fragment antigen-binding) region originates from a humanized murine antibody. Moreover, the antibody is unlikely to interfere with the immune function of CD4, because this function is mediated through domain 1 of the receptor, an assumption that was confirmed by *in vitro* studies in human and monkey blood cells. *In vitro* tests also showed no induction of blood-cell apoptosis, and there was no depletion of CD4 cells in monkeys.

The investigators hope that their treatment approach will circumvent drug resistance, because the target is a human glycoprotein rather than a highly mutable viral protein. The Phase I dose-escalation study that is about to start will evaluate the safety and tolerability of a

1359-6446/01/$ – see front matter PII: S1359-6446(01)02009-8