

# Letters to the Editor

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## Is Low-Heat Shock Protein 70 a Primary or a Secondary Event in the Development of Atherosclerosis?

Letter to the Editor:

Pockley et al's landmark observation that serum heat shock protein (Hsp) 70 levels predict the development of atherosclerosis<sup>1</sup> prompts the question whether Hsp 70 is low as a result of atherosclerosis or the primary event leading to atherosclerosis. We believe an argument can be made for either a primary or a secondary event, or even a combination of both. First, low Hsp 70, itself, should add to the vulnerability of the artery to stress because, indeed, Hsp 70 protects cellular elements from injury by reducing oxidation, inflammation, and apoptosis and by refolding damaged proteins.<sup>2</sup>

As a secondary event, Hsp 70 may be low as a result of decreased release of nitric oxide into the circulation caused by endothelial dysfunction. Nitric oxide normally has an oxidizing effect that increases Hsp 70 expression, additionally, studies show that blocking nitric oxide release reduces Hsp expression.<sup>3</sup> Thus, low levels of Hsp 70 in subjects with progressive atherosclerosis could be the result of primary endothelial dysfunction.

However, reduced Hsp 70 may be a primary event in the development of atherosclerosis. Hsp 70 recently has been found to be markedly low in the skeletal muscle of individuals with type 2 diabetes and moderately low in the nondiabetic identical twin with a diabetic co-twin.<sup>4,5</sup> Furthermore, in a study comparing 5600 genes of nondiabetic subjects with insulin-resistant diabetic subjects, Hsp 70 was one of only 17 genes that were significantly lower in individuals with diabetes.<sup>6</sup> These studies suggest that reduced Hsp 70 may be a necessary event leading to the development of diabetes and subsequent atherosclerosis.

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### **Response: Is Low-Heat Shock Protein 70 a Primary or a Secondary Event in the Development of Atherosclerosis?**

We thank Dr Philip Hooper and Dr Joanna Hooper for their comments on our recent article.<sup>1</sup> Their question as to whether Hsp70 levels are low as a result of atherosclerosis or whether low

Hsp70 levels are the primary event leading to atherosclerosis is clearly an important one. Our previous study in which we reported there to be no relationship between serum levels of Hsp70 and the presence of atherosclerosis in subjects with established hypertension, as determined on the basis of carotid intima-media thickness,<sup>2</sup> would argue against the proposition that Hsp70 levels are low as a result of atherosclerosis. However, another study has reported Hsp70 levels to be lower in patients at the time of diagnosis of coronary artery disease by coronary angiography.<sup>3</sup> In Zhu et al's study, individuals exhibiting Hsp70 levels below the median had twice the risk of coronary artery disease than individuals with levels above the median, and disease severity (number of diseased vessels) was also inversely associated with circulating Hsp70 levels.<sup>3</sup>

As the Hoopers indicate, Hsp70 is a cytoprotective molecule, and a deficiency in its presence might increase the vulnerability of cardiovascular tissue to environmental and physical stressors. The majority of work in this area has focused on Hsp70 as an intracellular molecule, and little is known about its ability to protect cells when present in the extracellular environment. One study has addressed this issue and demonstrated that extracellular Hsp70 protects stressed aortic cells in culture by a mechanism that appears to involve cell surface binding, but not internalization.<sup>4</sup> However, the mechanism by which this effect is achieved has never been elucidated.<sup>4</sup>

Key issues will be to identify the source of Hsp70 in the peripheral circulation and to determine the mechanism by which Hsp70 is released and whether extracellular levels reflect intracellular levels. Were the origin of circulating Hsp70 to be the endothelium, then it might be reasonable to assume that levels are influenced by endothelial dysfunction, either directly or via the compromised generation of nitric oxide. However, Hsp70 induces nitric oxide production from macrophages,<sup>5</sup> and if it is similarly capable of inducing nitric oxide production from endothelial cells, then it might be difficult to distinguish the primary event and evaluate its significance to endothelial dysfunction.

Another mechanism by which Hsp70 might modify the establishment and/or progression of atherosclerosis is via an anti-inflammatory effect. Intracellular Hsp70 has been shown to attenuate inflammatory responses, because elevating intracellular levels of Hsp70 in the vasculature reduces leukocyte adhesion at inflammatory sites.<sup>6</sup> Immunization with Hsp70 has also been shown to prevent disease in experimental models of autoimmunity.<sup>7–9</sup> Although purely speculative at this time, it might be that elevated serum Hsp70 levels reflect an antiinflammatory and/or an antiatherogenic state.

In summary, the presence of heat shock proteins in the peripheral circulation is a relatively recently reported phenomenon<sup>10,11</sup> and its biological and clinical significance has yet to be elucidated. The basis for the apparent relationship between high-serum Hsp70 levels and protection against atherosclerosis is currently unknown, and further work is required to understand the influence of extracellular Hsp70 on the pathogenesis of inflammatory disease.

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