CRP as a Clinical Marker

Compelling evidence exists to support CRP’s designation as a powerful independent predictor of future cardiovascular risk. CRP predicts cardiovascular risk in a wide variety of clinical settings, including men and women without overt cardiovascular disease. Patients with stable angina or presenting with acute coronary syndromes, postmyocardial infarction patients, and those with the metabolic syndrome. Furthermore, CRP predicts not only incident myocardial infarction and cardiovascular death, but also the risk of ischemic stroke, sudden cardiac death, incident peripheral artery disease, and restenosis after percutaneous coronary intervention. In primary prevention, CRP confers additional prognostic value at all levels of Framingham risk and at all levels of the metabolic syndrome and blood pressure. In head-to-head comparisons with LDL cholesterol, CRP was found to be the stronger predictor of incident cardiovascular events. This robust association with future cardiovascular events has provided an analytic opportunity for CRP in clinical use. CRP is an appropriate marker because it has a long half-life, CRP levels remain stable over time without exhibiting circadian variability, and fasting blood samples are not required. Currently, CRP levels <1 mg/L, 1 to 3 mg/L, and >3 mg/L are used to denote low- intermediate-, and high-risk groups. However, differentiation between the different forms of CRP is not available at the present time. Though unknown, the findings by Khreiss et al raise the possibility that patients with more monomeric CRP or CRP with a tendency to become modified more readily may exhibit a greater proinflammatory phenotype and hence be at even greater risk for adverse cardiovascular events.

CRP testing provides prognostic information, but does it also provide a target for treatment to enhance survival, like LDL does? Lifestyle modification provides the first opportunity to modify CRP levels because obesity, smoking, diabetes, and lack of exercise are associated with elevated CRP levels. Furthermore, strong evidence has accumulated to suggest that the benefits of risk-reduction strategies, including aspirin and statins, are markedly enhanced in patients with elevated CRP. However, there is no evidence yet that lowering CRP will necessarily lower cardiac risk. Whether pharmacological treatment of patients who only have elevated CRP is warranted will require further trials. Currently, the Justification for the Use of statins in Primary prevention: an Intervention Trial Evaluating Rosuvastatin (JUPITER) is being conducted to determine if the treatment of otherwise healthy individuals with low levels of LDL but high levels of CRP will benefit from statin therapy. However, by uncovering the structural and molecular mechanisms by which CRP interacts with the endothelium, new therapeutic targets may be uncovered. For example, agents interfering with the conversion of pentameric CRP to the more active monomeric form may serve to limit CRP’s proinflammatory effects. Agents that destabilize monomeric CRP may also serve to limit its detrimental effect. Though the concept is in the preliminary stage, as Khreiss et al demonstrated, blocking CRP receptors may serve as a novel strategy to limit endothelial cell activation. However, further studies are required to confirm the presence of and identify the major binding sites for monomeric CRP on endothelial cells.

CRP Genetics

Baseline levels of CRP show a clear heritability of approximately 40% in studies of families. Currently, 3 poly-
morphisms in the CRP gene that are associated with changes in CRP level have been documented. By identifying genetic variations in the CRP gene, at-risk genotypes may be deciphered, providing additional information for overall risk assessment. Such genotype-specific risk categories may identify individuals who have relatively low serum CRP levels yet display an enhanced proinflammatory phenotype. Perhaps these higher-risk individuals will be found to have a polymorphism that decreases the stability of the pentameric structure, thereby promoting monomeric CRP formation, or alternatively, one that increases the stability of monomeric CRP binding to the cell membrane, thereby resulting in enhanced endothelial cell activation. Thus, the study by Khreiss et al may provide the stimulus to uncover such new genetic variants.

**CRP as a Mediator of Atherosclerosis**

Human recombinant CRP, at concentrations known to predict vascular disease, elicits a multitude of effects on endothelial biology favoring a proinflammatory and proatherosclerotic phenotype. In vitro experiments reveal that CRP potently downregulates eNOS transcription and destabilizes eNOS mRNA, resulting in decreased release of basal and stimulated NO, a key endothelium-derived relaxing factor. In a synchronous fashion, CRP has been shown to stimulate endothelin-1 and interleukin-6 release from endothelial cells; upregulate adhesion molecules such as intercellular adhesion molecule-1, vascular cell adhesion molecule-1, and E-selectin; and stimulate the release of monocyte chemotactic protein-1, a chemokine that facilitates leukocyte transmigration. By inhibiting NO production, CRP facilitates endothelial cell apoptosis and blocks angiogenesis. Furthermore, CRP augments CD14-induced endothelial cell activation; potently upregulates nuclear factor-κB, a key nuclear factor that facilitates the transcription of numerous proatherosclerotic genes; and inhibits bone marrow–derived endothelial progenitor cell survival and differentiation. Endothelial progenitor cells have been suggested to play an important role in postnatal neovascularization, and the ability of CRP to inhibit progenitor cells may be an important mechanism inhibiting compensatory angiogenesis in chronic ischemia.

CRP also has direct proatherogenic effects in vascular smooth muscle cells. Recent evidence suggests that CRP directly upregulates angiotensin type 1 receptor in vascular smooth muscle cells in vitro and in vivo and stimulates vascular smooth muscle migration, proliferation, neointimal formation, and reactive oxygen species production. However, we have also shown that CRP upregulates complement-inhibitory proteins and protects endothelial cells from complement-mediated cell injury. This suggests that a balance of proatherogenic and antiatherogenic
effects of CRP on the vessel wall may be important in the development of atherosclerosis.

Recently, the question of whether the accumulating in vitro data have direct clinical relevance has been debated, given that the CRP concentrations used are typically in excess of 5 mg/L. These in vitro experiments used native, pentameric CRP. Though it is likely that circulating levels of CRP do not truly reflect the tissue concentrations of CRP, the study by Khreiss et al adds support to this theory. CRP must undergo a structural change on cell membrane binding for endothelial activation, and this active component is deposited in the wall of the blood vessel, where it can promote atherogenesis. Khreiss et al show that monomeric CRP was able to induce endothelial activation within 4 hours, whereas with native CRP, similar results were not obtained until 24 hours later, because monomeric CRP actions were based on a tissue rather than a serum environment. Because the effects of native CRP are dependent on an unidentified serum cofactor, a cofactor that may not be present at physiological levels in cell culture, in vitro concentrations of CRP may need to be higher to compensate for this difference.

With the development of a human CRP-transgenic (CRP-tg) mouse, an animal model is now available to determine the role CRP plays in vivo. Two studies recently published in Circulation revealed that expression of human CRP in mice actively promoted adverse cardiovascular processes. First, human CRP created a prothrombotic phenotype, as evidenced by higher rates of thrombotic occlusion after arterial injury. Second, by crossing CRP-tg mice with apoE/H11002, these CRP-tg/apoE−/− mice, CRP was shown to be an active player in atherogenesis in vivo. These CRP-tg/apoE−/− mice displayed accelerated aortic atherosclerosis, which was associated with increased complement deposition and elevated expression of angiotensin type 1 receptor, vascular cell adhesion molecule-1, and collagen within the lesions. Further studies using these CRP-tg mice will no doubt confirm the in vitro results implicating CRP with endothelial dysfunction, as evidenced by impaired NO production and enhanced endothelin-I release. Furthermore, it will provide a model to assess new therapeutic strategies aimed at decreasing CRP levels and to determine whether this strategy has an effect on plaque initiation, progression, and rupture.

Conclusion
The CRP–atherosclerosis story continues to unravel. With every new discovery, more questions emerge. Now that CRP appears to play the role of a participant and not just that of an indicator of atherosclerosis, uncovering the molecular mechanisms behind this interaction is more important. The finding that monomeric CRP is a prerequisite for endothelial cell activation is a first step in this direction. Firmly identifying how the pentameric structure unravels, what the actual CRP receptor on the surface of endothelial cells is, what the molecular signals it initiates are, and how these pathways could be modulated may uncover a novel target to prevent and treat atherosclerotic vascular disease.

References


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