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Jupiter to Earth: CRP Promotes Atherothrombosis

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Recently, the Justification for the Use of Statins in Primary Prevention: An Intervention Trial Evaluating Rosuvastatin (JUPITER), a landmark primary-prevention trial, was published.¹ The major objective of JUPITER was to investigate whether treatment with rosuvastatin, 20 mg daily, compared to placebo, would decrease the rate of first major cardiovascular events in healthy subjects with normal low-density lipoprotein cholesterol (LDL-C) but elevated C-reactive protein (CRP) levels. JUPITER was a randomized, double-blind, placebo-controlled, multicenter trial conducted at 1315 sites in 26 countries. Men, 50 years of age or older and women, 60 years of age or older, were eligible for the trial if they did not have a history of cardiovascular disease and if, at the initial screening visit, they had an LDL-C level of <130 mg/dL (3.4 mmol/L) and a high-sensitivity CRP level of >2.0 mg/L. Eligible subjects were randomly assigned in a 1:1 ratio to receive either rosuvastatin, 20 mg daily, or matching placebo. The primary outcome was the occurrence of a first major cardiovascular event, defined as nonfatal myocardial infarction, nonfatal stroke, hospitalization for unstable angina, an arterial revascularization procedure, or confirmed death from cardiovascular causes.

This trial of 17,802 individuals was stopped by the data and safety monitoring board after a median 1.9 years of the proposed 5 years of follow up because of a significant reduction in the primary end point in the rosuvastatin group. Treatment with rosuvastatin significantly reduced the primary composite end point 44% compared to placebo. There was also a significant 20% reduction in total mortality. The only significant adverse reaction was an increase in the frequency of physician-reported diabetes (3.0% versus 2.4%) and a significant increase in glycosylated hemoglobin (HbA1c) levels, with no difference in fasting plasma glucose levels. For participants who had elevated levels of high-sensitivity CRP but who were nonsmokers, were not overweight (had a body-mass index <25 kg/m²), did not have the metabolic syndrome, had a calculated Framingham risk score of 10% or less, or had an LDL-C level of 100 mg/dL (2.6 mmol/L) or lower, the observed relative reductions in the hazard ratio associated with rosuvastatin for the primary end point were similar to those in higher-risk groups. Most interestingly, among patients with no other major cardiovascular risk factor other than increased age, rosuvastatin therapy resulted in a significant 37% reduction in cardiovascular events. Whereas rosuvastatin reduced LDL-C by 50%, the reduction in cardiovascular events in JUPITER was almost twice that predicted based on LDL-C reduction on the basis of previous statin trials. It is important to point out that CRP levels were significantly reduced by 37% to a median value of 2.2 mg/L.

Measurement of high-sensitivity CRP, the prototypic bio-marker of inflammation that independently predicts future vascular events, improves global classification of risk, regardless of the LDL-C level.² Previously, we have shown in a prospective study that the reduction in CRP levels with statins is a class effect.³ In the PROVE-IT study,⁴ patients with acute coronary syndromes (ACS) with concomitant reduction in LDL-C <70 mg/dL and CRP <2 mg/L had the greatest benefit. To date, however, this is the first prospective outcome trial that has directly

addressed the question of whether apparently healthy persons with levels of LDL-C below current treatment thresholds but with elevated levels of high-sensitivity CRP benefit from statin therapy. Findings from the study indicate that we can no longer assume that patients with low cholesterol are at low risk. Indeed, patients with low cholesterol and no other Framingham risk score feature but high CRP benefited greatly from statin therapy. While we are not able to discern how much the CRP or LDL lowering each contributed to the reduction in cardiovascular events, it is important to note that other prospective trials with larger doses of statins (SEARCH, IDEAL, A to Z)⁵⁻⁷ do not show greater reductions in cardiovascular events in spite of greater LDL-C reduction.

The new Adult Treatment Panel III (ATP III) guidelines will now need to consider these convincing results by recommending CRP testing to intermediate-risk patients so that we can better identify candidates at greater risk. Increased CRP levels integrate a myriad of metabolic abnormalities, such as increased adiposity, diabetes, smoking, end-stage renal disease, hypertension, metabolic syndrome, etc. If high-sensitivity CRP is recommended as a screening test, it is important that the test be performed on two occasions at least a week apart, with the patient being in a steady state. If levels exceed 10 mg/L, then one should search for a nidus of inflammation and then repeat the test once the inflammatory episode has resolved.

How does the vascular biologist interpret the JUPITER study with respect to the role of CRP in atherothrombosis? The most important message is that in patients with high CRP and no other major risk factor, statin therapy resulted in a significant benefit. Thus, at minimum, these data further validate the hypothesis that CRP is an active participant in atherothrombosis and that lowering CRP is beneficial. Many groups have shown that CRP promotes potent proatherogenic effects in endothelial cells, smooth muscle cells, platelets, and monocyte macrophages *in vitro*.^{8,9} More importantly, there is emerging evidence from different studies that *in vivo* CRP administration exerts these effects in appropriate animal models and in human infusion studies. Griselli et al.^{8,9} showed that administration of human CRP in experimental acute myocardial infarction produced by coronary artery ligation reproducibly enhanced infarct size by ~40%. Gill et al.^{8,9} also showed that adult rats subjected to middle cerebral artery occlusion and then treated with human CRP similarly developed significantly larger cerebral infarcts compared to human serum albumin treatment. We have shown that *in vivo* administration of carefully prepared human CRP (that has biological effects in Toll-like receptor knockdown endothelial cells¹⁰) compared to human serum albumin significantly increased oxidized LDL uptake and intracellular cholesterol ester accumulation in macrophages, activated matrix metalloproteinase-9 (MMP-9), and upregulated nicotinamide adenine dinucleotide phosphate (NADPH) oxidase, resulting in an increase in reactive oxygen species and tissue factor release.^{8,9} Furthermore, in 7 male volunteers Bisio et al.^{8,9} showed that human CRP infusion resulted in an increase in plasma CRP, and subsequently both biomarkers of inflammation and coagulation were activated.

Thus, it behooves researchers in this area to focus on *in vivo* studies in appropriate animal models and in patients in whom high CRP conveys greater risk, such as metabolic syndrome, acute coronary syndromes, and the like. In rats, the Pepys group¹¹ has shown that a specific CRP inhibitor (bis-phosphocholine) decreases myocardial infarct size, lending further credence to the hypothesis that CRP promotes atherothrombosis. Although the perfect strategy would be of a drug that selectively inhibits CRP and not LDL-C to prove the point that CRP contributes to atherothrombosis, results of clinical trials with a CRP inhibitor are not available. Strategies targeting CRP specifically, such as antisense technology, will ultimately confirm the evolving and exciting role of CRP in atherothrombosis. Another reasonable strategy will be a trial in patients with metabolic syndrome with elevated CRP levels aimed at reducing CRP and not LDL-C, such as with the peroxisome proliferator-activated receptor- γ (PPAR- γ) agonists (e.g., pioglitazone) that decrease CRP and reduce cardiovascular events in diabetics with

cardiovascular disease.^{12,13} Additionally, methotrexate therapy in patients with psoriasis will also prove instructive because the drug lowers CRP and these patients appear to be at increased risk for cardiovascular events.¹⁴ In the meantime, research efforts should be directed at further understanding the vascular effects of CRP, especially *in vivo*. In this regard, it is important to emphasize that the inhibition of endothelial nitric oxide synthase reported in 2002 has now been confirmed in animal models and in human volunteers following infusion of carefully prepared CRP, as manifested by impaired endothelial vasoreactivity.^{15,16}

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References

1. Ridker PM, Danielson E, Fonseca F, Genest J, Gotto A, Kastelein J, et al. Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. *N Engl J Med* 2008;359:2195–2207. [PubMed: 18997196]
2. Verma S, Szmitko PE, Ridker PM. C-reactive protein comes of age. *Nat Clin Pract Cardiovasc Med* 2005;2:29–36. [PubMed: 16265340]
3. Jialal I, Stein D, Balis D, Grundy SM, Adams-Huet B, Devaraj S. Effect of hydroxymethyl glutaryl coenzyme a reductase inhibitor therapy on high sensitive C-reactive protein levels. *Circulation* 2001;103:1933–1935. [PubMed: 11306519]
4. Ridker RM, Cannon CP, Morrow D, Rifai N, Rose LM, McCabe CH, et al. Pravastatin or Atorvastatin Evaluation and Infection Therapy-Thrombolysis in Myocardial Infarction 22 (PROVE IT-TIMI 22) investigators. C-reactive protein levels and outcomes after statin therapy. *N Engl J Med* 2005;352:20–28. [PubMed: 15635109]
5. Bowman L, Armitage J, Bulbulia R, Parish S, Collins R. SEARCH Study Collaboration Group. Study of the Effectiveness of Additional Reductions in Cholesterol and Homocysteine (SEARCH): Characteristics of a randomized trial among 12064 myocardial infarction survivors. *Am Heart J* 2007;154:815–823. [PubMed: 17967584]
6. Pedersen TR, Faergeman O, Kastelein JJ, Olsson AG, Tikkanen MJ, Holme I, et al. Incremental Decrease in End Points Through Aggressive Lipid Lowering (IDEAL) Study Group. High-dose atorvastatin vs usual-dose simvastatin for secondary prevention after myocardial infarction: the IDEAL study: a randomized controlled trial. *JAMA* 2005;294:2437–2445. [PubMed: 16287954]
7. de Lemos JA, Blazing MA, Wiviott SD, Lewis EF, Fox KA, White HD, Rouleau JL, Pedersen TR, Gardner LH, Mukherjee R, Ramsey KE, Palmisano J, Bilheimer DW, Pfeffer MA, Califf RM, Braunwald E. Early intensive vs a delayed conservative simvastatin strategy in patients with acute coronary syndromes: phase Z of the A to Z trial. *JAMA* 2004;292:1307–1316. [PubMed: 15337732]
8. Verma S, Devaraj S, Jialal I. Is C-reactive protein an innocent bystander or proatherogenic culprit? C-reactive protein promotes atherothrombosis. *Circulation* 2006;113:2135–2151. [PubMed: 16671184]
9. Devaraj S, Singh U, Jialal I. The Evolving Role of CRP in Atherothrombosis. *Clin Chem*. 2008(in press)
10. Dasu MR, Devaraj S, Du Clos TW, Jialal I. The biological effects of CRP are not attributable to endotoxin contamination: evidence from TLR4 knockdown human aortic endothelial cells. *J Lipid Res* 2007;48:509–512. [PubMed: 17158793]
11. Pepys MB, Hirschfield GM, Tennent GA, Gallimore JR, Kahan MC, Bellotti V, et al. Targeting C-reactive protein for the treatment of cardiovascular disease. *Nature* 2006;440:1217–1221. [PubMed: 16642000]
12. Hanefeld M, Marx N, Pfitzner A, Baurecht W, Lubben G, Karagiannis E, et al. Anti-inflammatory effects of pioglitazone and/or simvastatin in high cardiovascular risk patients with elevated high sensitivity C-reactive protein. *J Am Coll Cardiol* 2007;49:290–297. [PubMed: 17239709]
13. Dormandy JA, Charbonnel B, Eckland D, Erdmann E, Massi-Benedetti M, Moules IK, et al. Secondary prevention of macrovascular events in patients with type 2 diabetes in the PROactive

Study (PROspective pioglitAzone Clinical Trial In macroVascular Events): a randomised controlled trial. *Lancet* 2005;366:1279–1289. [PubMed: 16214598]

14. Dasu MR, Devaraj S, Du Clos TW, Jialal I. The biological effects of CRP are not attributable to endotoxin contamination: evidence from TLR4 knockdown human aortic endothelial cells. *J Lipid Res* 2007;48:509–512. [PubMed: 17158793]
15. Jialal I, Verma S, Devaraj S. Inhibition of endothelial nitric oxide synthase by C reactive protein. *Clin Chem*. 2008
16. Bisioendial RJ, Kastelein JJ, Peters SL, Levels JH, Birjmohun R, Rotmans JI, Hartman D, Meijers JC, Levi M, Stroes ES. Effects of CRP infusion on endothelial function and coagulation in normocholesterolemic and hypercholesterolemic subjects. *J Lipid Res* 2007;48:952–960. [PubMed: 17259597]