Plasma Concentration of C-Reactive Protein and Risk of Developing Peripheral Vascular Disease

Paul M. Ridker, MD; Mary Cushman, MD; Meir J. Stampfer, MD; Russell P. Tracy, PhD; Charles H. Hennekens, MD

Background—Among apparently healthy men, elevated levels of C-reactive protein (CRP), a marker for systemic inflammation, predict risk of myocardial infarction and thromboembolic stroke. Whether increased levels of CRP are also associated with the development of symptomatic peripheral arterial disease (PAD) is unknown.

Methods and Results—Using a prospective, nested, case-control design, we measured baseline levels of CRP in 144 apparently healthy men participating in the Physicians’ Health Study who subsequently developed symptomatic PAD (intermittent claudication or need for revascularization) and in an equal number of control subjects matched on the basis of age and smoking habit who remained free of vascular disease during a follow-up period of 60 months. Median CRP levels at baseline were significantly higher among those who subsequently developed PAD (1.34 versus 0.99 mg/L; P= .04). Furthermore, the risks of developing PAD increased significantly with each increasing quartile of baseline CRP concentration such that relative risks of PAD from lowest (referent) to highest quartile of CRP were 1.0, 1.3, 2.0, and 2.1 (P_trend=.02). Compared with those with no clinical evidence of disease, the subgroup of case patients who required revascularization had the highest baseline CRP levels (median=1.75 mg/L; P=.04); relative risks from lowest to highest quartile of CRP for this end point were 1.0, 1.8, 3.8, and 4.1 (P_trend=.02). Risk estimates were similar after additional control for body mass index, hypercholesterolemia, hypertension, diabetes, and a family history of premature atherosclerosis.

Conclusions—These prospective data indicate that among apparently healthy men, baseline levels of CRP predict future risk of developing symptomatic PAD and thus provide further support for the hypothesis that chronic inflammation is important in the pathogenesis of atherothrombosis. (Circulation. 1998;97:425–428.)

Key Words: C-reactive protein n inflammation n claudication n arteries n atherosclerosis n risk factors

C laudication due to peripheral arterial vascular disease is a common condition, affecting 2% to 5% of the United States population older than 50 years of age. Among patients with severe claudication, chronic lower extremity ischemia can lead to recurrent infection, need for surgical revascularization, and limb loss. In general, risk factors for peripheral arterial disease (PAD) are similar to those for coronary heart disease. However, as with coronary disease, many patients develop symptomatic claudication without such factors.

Recent studies suggest that low-grade inflammation is present among patients at risk for future atherothrombotic disease, at least in the coronary and cerebral circulations. Specifically, elevated levels of C-reactive protein (CRP), a marker for systemic inflammation, have been found among individuals with stable and unstable angina who are at risk for future myocardial infarction or sudden death, elderly patients at risk for symptomatic coronary heart disease, those at high risk for coronary death, and apparently healthy men at risk for first-ever myocardial infarction or stroke. As such, it has been hypothesized that CRP may provide a molecular marker of the underlying severity of preclinical atherosclerosis. Currently, few data are available relating CRP to future risk of developing symptomatic PAD. We therefore sought to determine whether increased levels of CRP among apparently healthy men are also associated with increased risk of developing symptomatic claudication or need for peripheral arterial revascularization.

Methods

The study population consisted of apparently healthy men participating in the Physicians’ Health Study (PHS), a randomized, double-blind, placebo-controlled trial of aspirin and β-carotene in the primary prevention of heart disease and cancer conducted among 22,071 US male physicians aged 40 to 84 years. Participants had no prior history of cardiovascular disease or cancer and were randomly assigned to one of four treatments: 325 mg of aspirin on alternate days, 50 mg of β-carotene on alternate days, both, or neither. Before randomization, participants were asked to provide baseline blood samples; the proce-
TABLE 1. Baseline Characteristics of Study Participants

<table>
<thead>
<tr>
<th></th>
<th>Control Subjects (N=144)</th>
<th>Case Subjects (N=144)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>62.9±9.5</td>
<td>62.9±9.5</td>
<td>MC</td>
</tr>
<tr>
<td>Smoking status, %</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>30.1</td>
<td>30.1</td>
<td></td>
</tr>
<tr>
<td>Past</td>
<td>42.7</td>
<td>42.7</td>
<td>MC</td>
</tr>
<tr>
<td>Current</td>
<td>27.3</td>
<td>27.3</td>
<td></td>
</tr>
<tr>
<td>Body mass index, kg/m²</td>
<td>24.6±2.8</td>
<td>24.9±3.1</td>
<td>.3</td>
</tr>
<tr>
<td>Blood pressure, mm Hg</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Systolic</td>
<td>131.4±12.4</td>
<td>133.7±13.8</td>
<td>.1</td>
</tr>
<tr>
<td>Diastolic</td>
<td>79.9±6.6</td>
<td>80.6±7.2</td>
<td>.4</td>
</tr>
<tr>
<td>History of hyperlipidemia, %</td>
<td>8.5</td>
<td>10.2</td>
<td>.7</td>
</tr>
<tr>
<td>Diabetes, %</td>
<td>4.2</td>
<td>10.4</td>
<td>.04</td>
</tr>
<tr>
<td>Family history of premature atherosclerosis, %</td>
<td>9.2</td>
<td>16.7</td>
<td>.06</td>
</tr>
</tbody>
</table>

MC indicates matching criteria.

TABLE 2. Relative Risks of Developing Future Peripheral Arterial Disease According to Baseline Level of C-Reactive Protein

<table>
<thead>
<tr>
<th>Quartile of C-Reactive Protein (range, mg/L)</th>
<th>1 (&lt;0.55)</th>
<th>2 (0.55-0.99)</th>
<th>3 (1.0-2.1)</th>
<th>4 (&gt;2.1)</th>
<th>P_{\text{rand}}</th>
</tr>
</thead>
<tbody>
<tr>
<td>Case subjects (n=144), n (%)</td>
<td>23 (16)</td>
<td>29 (20)</td>
<td>45 (31)</td>
<td>47 (33)</td>
<td></td>
</tr>
<tr>
<td>Control subjects (n=144), n (%)</td>
<td>37 (26)</td>
<td>35 (24)</td>
<td>36 (25)</td>
<td>36 (25)</td>
<td></td>
</tr>
<tr>
<td>Crude analysis*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Relative risk</td>
<td>1.0</td>
<td>1.3</td>
<td>2.0</td>
<td>2.1</td>
<td>.02</td>
</tr>
<tr>
<td>95% CI</td>
<td>...</td>
<td>0.7-2.7</td>
<td>1.0-4.0</td>
<td>1.1-4.1</td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>...</td>
<td>0.4</td>
<td>0.04</td>
<td>0.03</td>
<td></td>
</tr>
<tr>
<td>Adjusted analysis†</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Relative risk</td>
<td>1.0</td>
<td>1.5</td>
<td>2.3</td>
<td>2.2</td>
<td>.02</td>
</tr>
<tr>
<td>95% CI</td>
<td>...</td>
<td>0.7-3.3</td>
<td>1.1-4.9</td>
<td>1.1-4.8</td>
<td></td>
</tr>
<tr>
<td>P</td>
<td>...</td>
<td>0.3</td>
<td>0.3</td>
<td>0.04</td>
<td></td>
</tr>
</tbody>
</table>

*Case-control pairs matched on age (±1 year), smoking status (current, past, never), and time since randomization.
†Adjusted analysis matched on age, smoking status, and time since randomization and controlled for body mass index, hypercholesterolemia, diabetes, a family history of premature atherosclerosis, and randomized aspirin assignment.

Blood specimens were analyzed in blinded pairs with the position of the patient's specimen varied at random to reduce the possibility of systematic bias and decrease interassay variability. The mean coefficient of variation was <.05.

Because CRP values are skewed toward higher values, median plasma concentrations were computed, and the significance of any differences in median values between case and control subjects was assessed by use of Wilcoxon rank sum test. Tests for trend were used to assess any relation of increasing CRP values with the risk of developing PAD after the study sample was divided into quartiles defined by the distribution of the control values. Adjusted estimates of risk were obtained by use of conditional logistic-regression models that accounted for the matching variables and also controlled for randomized treatment assignment, body mass index, diabetes, history of hypercholesterolemia, history of hypertension, and a family history of coronary artery disease. All probability values are two-tailed, and CIs are computed at the 95% level.

Results

Table 1 displays baseline clinical characteristics of the 144 initially healthy study subjects who subsequently developed claudication or underwent peripheral arterial revascularization during follow-up (case subjects) and of the 144 matched study participants who remained free of vascular disease (control subjects). Because case and control subjects were matched for age and smoking status, these variables are identical between groups. As expected, the prevalence of diabetes was higher among case subjects than control subjects. The median time to first report of PAD was 30 months after the collection of baseline blood samples.

Overall, median levels of CRP were significantly higher at baseline among those study participants who subsequently developed symptomatic PAD than among those who did not (1.34 versus 0.99 mg/mL; P=.04).

Table 2 displays the distribution of study subjects after the sample was divided into quartiles based on the distribution of CRP among control subjects. The relative risks of developing symptomatic PAD increased significantly with each increasing quartile of baseline concentration of CRP such that men in the highest quartile had a twofold increase in risk compared with men in the lowest quartile (relative risk, 2.1; 95% CI, 1.1 to 3.3).
provide evidence that this effect is not limited to the coronary artery disease cohort, elevated baseline levels of CRP were found to predict risk of future myocardial infarction and thromboembolic events.3 Thus, these data extend the role of CRP as a marker of vascular risk among otherwise healthy individuals and for those who remained free of vascular disease during follow-up (control subjects).

Median levels of CRP at baseline among study participants who subsequently developed intermittent claudication or who required peripheral arterial revascularization (case subjects) and for those who remained free of vascular disease during follow-up (control subjects).

4.1; \(P = .03\)). Specifically, the relative risks of developing future PAD from lowest (referent) to highest quartile of CRP at baseline were 1.0, 1.3, 2.0, and 2.1 (\(P_{\text{trend}} = .02\)). Analyses that further adjusted for body mass index, hypercholesterolemia, diabetes, and a family history of premature atherosclerosis had minimal impact on these relationships; after adjustment for these potential confounding factors, men in the highest quartile of CRP at baseline were found to have a risk of future PAD 2.2 times that of men in the lowest quartile (95% CI, 1.1 to 4.8; \(P = .04\)) (Table 2).

As shown in the Figure, median levels of baseline CRP were highest in the subgroup of 31 case subjects who required peripheral arterial revascularization in addition to developing intermittent claudication. Relative risks for this end point increased significantly from lowest to highest quartile of baseline CRP (relative risks, 1.0, 1.8, 3.8, and 4.1; \(P_{\text{trend}} = .02\)). After adjustment for body mass index, hypercholesterolemia, diabetes, and a family history of atherosclerosis, relative risks of revascularization from lowest to highest quartiles of baseline CRP were 1.0, 2.8, 8.6, and 7.1 (\(P_{\text{trend}} = .01\)).

In a prior report from the PHS,13 aspirin assignment was found to reduce the risk of peripheral arterial revascularization by 46%. In the current data, no statistically significant evidence of interaction was noted between randomized aspirin assignment and CRP among study participants who required peripheral revascularization.

**Discussion**

These prospective data indicate that among apparently healthy men, elevated baseline levels of CRP predict future risk of developing symptomatic PAD. In previous data from this cohort, elevated baseline levels of CRP were found to predict risk of future myocardial infarction and thromboembolic stroke.7 Thus, these data extend the role of CRP as a marker of vascular risk among otherwise healthy individuals and provide evidence that this effect is not limited to the coronary and cerebral circulations. Taken together, these observations and those from prior studies4-8 support the hypothesis that CRP may serve as a molecular marker for underlying systemic atherosclerosis.

Several hypotheses have been suggested as mechanisms by which CRP may increase atherosclerotic risk. For example, in addition to being a heptatically derived marker for systemic inflammation, it has been hypothesized that CRP may have procoagulant effects related to its ability to enhance expression of tissue factor.12 Experimental work also suggests that CRP can be found within endothelial vessel walls,13 that CRP avidly binds to human neutrophils,14 and that CRP can induce complement activation.15 Furthermore, recent data suggest that CRP and CRP peptides may be involved in processes related to shedding of some cellular adhesion molecules,16 an intriguing finding because these molecules play an important role in the adhesion and transmigration of leukocytes across the endothelial wall, an important step in the initiation of atherosclerosis. It has also been hypothesized that low-grade inflammation detected by CRP reflects evidence of chronic infection. In this regard, some cross-sectional and case-control studies have reported elevated antibody titers directed against *Chlamydia pneumoniae*, *Helicobacter pylori*, and cytomegalovirus among those with prevalent heart disease.17 Finally, it is possible that the association of CRP with vascular risk is the result of cytokines such as interleukin-6 that promote leukocyte adhesion and stimulate CRP production.18

In contrast to findings from this cohort for myocardial infarction,3 we found no evidence in these data of a statistically significant interaction between inflammation, aspirin, and risks of PAD. However, because data from the PHS suggest that aspirin reduces the need for peripheral arterial surgery but not intermittent claudication,13 this analysis was limited to those 31 case subjects who required revascularization. Thus, the power to detect a statistically significant interaction in these data is insufficient to exclude a true positive effect.

In summary, these prospective data indicate that baseline level of CRP predicts risk of future PAD. Moreover, the highest levels of CRP were found among those study participants who required surgical revascularization in addition to developing intermittent claudication. Thus, these data further support the hypothesis that CRP may serve as a molecular marker for underlying atherosclerosis and that higher levels correlate with greater extent of disease.3

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**References**


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