Impact of Prolonged Cyclooxygenase-2 Inhibition on Inflammatory Markers and Endothelial Function in Patients With Ischemic Heart Disease and Raised C-Reactive Protein
A Randomized Placebo-Controlled Study

Peter Bogaty, MD; James M. Brophy, MD, PhD; Martin Noel, MSc; Luce Boyer, RN; Serge Simard, MSc; Fernand Bertrand, BSc; Gilles R. Dagenais, MD

Background—The impact of cyclooxygenase (COX)-2 antagonist treatment on acute coronary risk is controversial. We investigated the effect of prolonged COX-2 inhibition on inflammatory profile and endothelial function in patients with ischemic heart disease and high serum C-reactive protein (CRP) values.

Methods and Results—In a double-blind study, 35 stable subjects on low-dose aspirin with ≥2 previous acute coronary events and 2 of 2 screening CRP values ≥2.0 mg/L were randomized to the COX-2 inhibitor rofecoxib (25 mg) or placebo daily for 6 months. Serum CRP, interleukin-6 (IL-6), P-selectin, matrix metalloproteinase-9 (MMP-9), and brachial artery endothelial function were evaluated. In the placebo group, CRP (median) was 3.16 mg/L (25% and 75% quartiles, 1.90 and 5.78 mg/L) at baseline and 4.22 mg/L (25% and 75% quartiles, 2.04 and 6.25 mg/L) at 6 months; in the rofecoxib group, CRP was 3.45 mg/L (25% and 75% quartiles, 2.08 and 5.78 mg/L) at baseline and 1.41 mg/L (25% and 75% quartiles, 1.17 and 4.81 mg/L) at 6 months (P=0.03). Rofecoxib compared with placebo also lowered IL-6 at 6 months (P=0.0002). There was a significant off-drug effect on CRP and IL-6 levels in the rofecoxib group 3 months after treatment (P=0.005 and P=0.009, respectively). Rofecoxib did not significantly affect P-selectin, MMP-9, and brachial artery vasoreactivity.

Conclusions—Prolonged COX-2 inhibition attenuates CRP and IL-6, does not modify P-selectin and MMP-9, and has no deleterious effect on endothelial function in stable patients with a history of recurrent acute coronary events and raised CRP. These results strengthen the rationale for evaluating the clinical benefit of COX-2 inhibition in patients with ischemic heart disease. (Circulation. 2004;110:934-939.)

Key Words: C-reactive protein ■ antiinflammatory agents, nonsteroidal ■ coronary disease ■ inflammation ■ interleukins

The effect of cyclooxygenase-2 (COX-2) antagonist treatment on acute coronary risk is a subject of continuing controversy. Although some studies have suggested a deleterious effect and others a neutral effect, their interpretation is problematic because of retrospective design, selection biases, uncontrolled nonuse of aspirin in higher-risk subsets, uncertainty about protection conferred by nonselective nonsteroidal antiinflammatory drugs in comparative trials, differences among COX-2 inhibitors, and possible play of chance when acute coronary event rates are very low. However, there are premises to support a beneficial effect of antiinflammatory therapy in patients with ischemic heart disease. The importance of inflammation in atherosclerosis is increasingly recognized. An enhanced inflammatory biological profile, as evidenced by raised serum inflammatory markers such as C-reactive protein (CRP), cytokines such as interleukin-6 (IL-6), the leukocyte-binding adhesion molecule P-selectin, and matrix metalloproteinase (MMP)-9, a macrophage enzyme believed to play a role in plaque instability, is associated with increased cardiovascular risk. An inflammatory biological profile is also associated with endothelial dysfunction in subjects with ischemic heart disease. In the last study, the spontaneous decrease in CRP in a few subjects was accompanied by an attenuation of endothelial dysfunction.

Thus, if inflammation is a significant contributor to ischemic heart disease and its clinical manifestations, antiinflammatory therapy could be beneficial. There are also specific premises for exploring the effect of COX-2 inhibition in atherosclerotic disease. COX-2 expression is increased in macrophage/foam cells and smooth muscle and endothelial...
cells of human atherosclerotic plaques\textsuperscript{14-16} and might stimulate plaque growth\textsuperscript{17} and activation of MMPs,\textsuperscript{18} favoring plaque instability. The few studies of COX-2 inhibition in experimental models of atherosclerosis, although not conclusive, suggest an antiatherosclerotic potential.\textsuperscript{19,20}

The effect of antiinflammatory agents such as aspirin and other COX inhibitors on mediators and markers of inflammation has been investigated in a few studies, but results are discordant. Aspirin had no effect on serum CRP in healthy volunteers,\textsuperscript{21,22} but it decreased CRP, IL-1b, IL-6, and macrophage colony-stimulating factor in patients with stable angina.\textsuperscript{23} Two recent studies with the COX-2 inhibitors celecoxib and rofecoxib in subjects with stable coronary artery disease had contradictory findings, a lowering of CRP and improved endothelial function in one study and a neutral effect in the other.\textsuperscript{24-25} However, all these studies were of short duration, and subjects had relatively low baseline CRP values.

Thus, the effects of antiinflammatory therapy on markers of inflammation and endothelial function in subjects with ischemic heart disease remain unclear, and long-term effects have not been investigated. To overcome some limitations of previous studies, we studied patients with a particularly pronounced inflammatory profile that was ensured (1) because they had a history of repetitive (\(\geq 2\)) acute coronary events\textsuperscript{26} and (2) because we required 2 of 2 screening CRP values to be in the higher range (\(\geq 2.0\) mg/L). These subjects were randomized to treatment with the COX-2 inhibitor rofecoxib 25 mg daily or placebo for 6 months. We evaluated serum values of CRP, IL-6, P-selectin, and MMP-9 and endothelial function. We hypothesized that the treatment effect would be a significant reduction in CRP at 6 months.

**Methods**

**Patient Selection**

We undertook a systematic review of hospital charts between 1996 and 2001, searching for patients \(\leq 72\) years of age with a well-documented history of \(\geq 2\) acute coronary events (myocardial infarction or unstable angina with a minimum of 1 infarction). The age criterion was arbitrarily set to realistically balance recruitment need with the greater risk of undesirable effects with prolonged COX-2 inhibitor therapy in an older cohort. The diagnosis of myocardial infarction required characteristic prolonged (\(\geq 30\) minutes) symptoms and creatine kinase (CK) elevation more than twice the upper normal limit (or CK-MB \(\geq 15\) U/L or \(\geq 5\%\) of total CK). Unstable angina required characteristic symptoms, either new in onset or a sharp and significant change in the pattern of established angina, appearing at rest or on minimum exertion and requiring hospitalization. The last acute coronary event must have occurred \(\geq 3\) months previously. Exclusion criteria were the following: uncontrolled hypertension (\(\geq 160/95\) mm Hg); serum creatinine \(\geq 160\) \(\mu\)mol/L or creatinine clearance \(< 40\) mL/min; left ventricular ejection fraction \(< 35\%\); history of congestive heart failure; history of cancer (unless \(\geq 5\) years in clinical remission) or any chronic debilitating condition; bleeding diathesis or clinically significant hemorrhagic episode; gastric or duodenal ulcer \(< 6\) months; warfarin use; hepatic dysfunction (AST, ALT, GGT, or bilirubin \(\geq 2\) times the upper normal limit); and known allergy to or intolerance of nonsteroidal antiinflammatory drugs. Patients meeting the eligibility criteria were invited to participate in this study if these conditions were met: (1) absence of any chronic or recent (\(\leq 1\) month) clinically significant infectious or inflammatory condition, trauma, vaccination, or invasive medical or dental procedure; (2) absence of any condition or context in which use of antiinflammatory medications was considered likely during the study; and (3) 2 of 2 screening serum CRP values \(> 2.0\) mg/L taken 14 days apart. The rationale for obtaining 2 screening values was to ensure a more stable estimate of patients’ CRP status, as recently recommended.\textsuperscript{26} The cutoff value for CRP was set at 2 mg/L to recruit a cohort with higher-range CRP values that would be optimal for detecting a treatment effect.

The hospital ethics committee approved the study, and all patients gave written informed consent.

**Study Procedure**

Thirty-five patients meeting the clinical and CRP eligibility criteria were randomized in double-blind fashion and 1:1 ratio to either 25 mg rofecoxib (VIOXX) or matching placebo daily for 6 months. The daily dosage of 25 mg has been shown sufficient to inhibit COX-2 activity in the range achieved by most antiinflammatory agents in standard doses\textsuperscript{27}; a 6-month treatment period was chosen to be more certain of not missing a treatment effect. All subjects were given enteric-coated aspirin 160 mg daily. All serum samples were obtained in mid morning in the nonfasting state at baseline (just before study drug was given); at 1, 3, and 6 months of study drug treatment; and 1 and 3 months after cessation of study medication. Serum was immediately centrifuged at 3500 rpm for 15 minutes; serum was placed in aliquots and promptly frozen at \(-80^\circ\)C. All measurements were performed in a single batch. CRP was measured with the N Latex CRP monoaassay (interassay reproducibility, 3.6\% to 4.4\%; sensitivity, 0.18 mg/L) using the Behring Nephelometer 100 Analyzer (Dade Behring). IL-6 was measured by a quantitative sandwich enzyme immunoassay (Roche Diagnostics GmbH; intra-assay variance \(< 8\%\); P-selectin and MMP-9 were measured with quantitative sandwich immunoassays (R&D Systems; intra-assay variabilities, 5\% and 2\%, respectively). Patients were regularly monitored for symptoms of study drug intolerance; electrolyte and creatinine status; blood pressure; fluid retention; concomitant medications, including nonprescription drugs; and aggravation of cardiac symptoms. If any clinically significant infectious or inflammatory condition occurred, blood sampling was either postponed to have a 10-day symptom- and event-free period, if still within 14 days of protocol time points, or not performed. If antiinflammatory treatment was necessary, it had to stopped 10 days before blood sampling. The protocol called for definitive withdrawal from the study in case of aggravation of cardiac symptoms, occurrence of unstable angina or myocardial infarction, occurrence of any significant undesired symptoms likely attributed to or rendering problematic the continuation of study medication, or the need for antiinflammatory medication for \(> 10\) consecutive days.

**Endothelial Function Testing**

Subjects underwent a baseline test of endothelial function, and a second test was performed at the end of the 6-month period of study drug administration. If study drug had to be stopped before 6 months, this latter test was performed at that time if the patient was on study drug treatment for \(\geq 3\) months and had no inflammatory condition within 10 days.

Endothelial function testing was performed in subjects after an overnight fast, between 7 and 8 AM, after a 15-minute rest in the supine position always in the same quiet dimly lit room. Subjects had to have consumed no caffeine or alcohol and were not to have smoked that morning. Vasoactive and antianginal medications were withheld 24 hours before testing.

At each test, a 3-lead ECG was applied with continuous monitoring. The right brachial artery was imaged longitudinally just above the antecubital fossa. The same precise location was always used subsequently. Images were captured at the same time of the cardiac cycle (peak of R wave) with a high-resolution ultrasound apparatus (Hewlett Packard 7-MHz linear-array vascular probe coupled to a Hewlett Packard 5500 ultrasound machine). Still frames were recorded 1 per 5-second interval, with the frames of the first and last 5 seconds of the subsequence discarded. At the initial baseline, a set of 9 frames was recorded during the 45 seconds. A blood pressure cuff was then inflated on the upper arm to 220 mm Hg for 5 minutes.
Immediately after release of the cuff, the brachial artery was continuously imaged, and 1 minute after release of the cuff, a set of 12 images was recorded during the following 90 seconds. After a 12-minute rest, a repeat baseline of 9 images was recorded. Sublingual nitroglycerin 0.3 mg was administered, and 9 more images were recorded 3 minutes later.

Recorded images were analyzed by an experienced technician who was blinded to clinical data, was unaware of the order of the tests, and used dedicated image analysis software (Dynamic Endothelial Assessment V2.0, Vasometrix). The mean calibrated diameter of the artery segment was calculated from an average of 120 computerized diameter measurements per frame digitalized. The average of all frames of each subsequence was then computed. The percent changes in arterial caliber after flow-mediated vasodilation (FMV) and after nitroglycerin-mediated vasodilation (NMV) from their respective basal states were determined at the baseline visit and at 6 months.

**Statistical Analysis**

We determined that a sample size of 35 patients (assuming a 10% dropout rate) would be needed with \( n = 0.05 \) and a power of 90% to detect a 30% drug-lowering effect on the principal parameter of interest, CRP, from baseline to the measurement at 6 months of treatment. From our previous work, this sample size was also sufficiently powered to detect a treatment effect on endothelial function.\(^{28}\)

Results are expressed in percent for categorical variables and as mean±SEM for continuous variables except for CRP and IL-6, which had a skewed distribution and are represented as medians (25% and 75% quartiles) and were logarthimically transformed for analysis. Student’s \( t \) test was used to compare means, and the \( \chi^2 \) statistic was used to compare proportions. A 3-factor ANOVA model was used to analyze the serum markers of interest and brachial artery reactivity (FMV and NMV). The treatment factor was defined as the comparison between the placebo and rofecoxib groups; the subject factor, nested in the treatment factor, was analyzed as a random effect; the time factor considered data at baseline and at 6 months of treatment. A mixed-model analysis was performed with an interaction term between the fixed factors. Different statistical models were tested to obtain the best-fitting model. Comparisons of the adjusted Akaike’s information criterion for the different models were performed. To proceed with the ANOVA, we used a model with a variance component structure. A covariance repeated-measures ANOVA was performed to control for confounding variables. Univariate and multivariate normalities and variance assumptions were fulfilled. Relations between variables were analyzed with the Pearson correlation coefficient. The level of statistical significance was set at \( P<0.05 \). Analyses were based on the intention-to-treat principle. The statistical package SAS version 8.2 (SAS Institute Inc) was used.

**Results**

Of 295 clinically eligible patients, 174 patients were excluded, mostly because they lived too far away or declined to participate. The remaining 121 patients consented to CRP screening, and 61 of these patients (50.4%) had a first serum CRP >2 mg/L. Forty-six of these patients (35.4%) had a second CRP value >2 mg/L within 14 to 21 days, and 35 of them agreed to be randomized, 18 patients to rofecoxib and 17 patients to placebo. Two patients discontinued within 2 weeks of randomization (both on placebo; 1 voluntarily and 1 because of the occurrence of herpes zoster). One patient on rofecoxib had unstable angina 1 month after randomization; 1 patient withdrew after 3 months because of dyspepsia (on placebo); 1 patient had a myocardial infarction 1 month after the 6-month study drug treatment period (on placebo); 1 patient (on rofecoxib) developed a vasculitic purpura just before final blood sampling 3 months after treatment cessation; and 1 patient had a respiratory infection just before final blood sampling. The data from these patients were retained for analysis until their study withdrawal. One patient had an unaccountably very high CRP (47.9 mg/L) at 6 months, and this value was not retained. During the study, a diuretic was begun in 2 patients because of peripheral edema and increased blood pressure, respectively; a proton-pump inhibitor was begun in 4 patients because of dyspepsia; and an ACE inhibitor was begun in 3 patients because of increased blood pressure. Adjustment for these medications did not affect the results. Serum creatinine levels remained stable in all patients during the study. Study drug compliance was 97.0±0.6%.

The 35 study patients (32 men, 3 women) were 58±1 years of age. They had a history of 1.7±0.1 myocardial infarctions and 0.9±0.2 episodes of unstable angina. The first and second qualifying (prebaseline) CRP values were 4.40 mg/L (25th and 75th percentiles, 2.90 and 7.60 mg/L) and 3.51 mg/L (25th and 75th percentiles, 2.50 and 6.30 mg/L), respectively (\( P=0.16 \)). Clinical characteristics of study patients on placebo and rofecoxib are shown in Table 1. The study group was relatively obese (body mass index, 32.2±0.8 kg/m\(^2\)). The rofecoxib and placebo groups were generally well matched, although there were 4 patients with diabetes in the former and none in the latter.

**Values are mean±SEM when appropriate.**

**TABLE 1. Baseline Clinical Characteristics of the 2 Study Groups**

<table>
<thead>
<tr>
<th></th>
<th>Rofecoxib (( n=18 ))</th>
<th>Placebo (( n=17 ))</th>
<th>( P )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>59±2</td>
<td>58±2</td>
<td>0.8</td>
</tr>
<tr>
<td>Male sex, n (%)</td>
<td>16 (88.8)</td>
<td>16 (94.1)</td>
<td>1.0</td>
</tr>
<tr>
<td>Hypertension, n (%)</td>
<td>6 (33.3)</td>
<td>10 (58.8)</td>
<td>0.2</td>
</tr>
<tr>
<td>Diabetes, n (%)</td>
<td>4 (22.2)</td>
<td>0</td>
<td>0.1</td>
</tr>
<tr>
<td>Hypercholesterolemia, n (%)</td>
<td>18 (100)</td>
<td>17 (100)</td>
<td>1.0</td>
</tr>
<tr>
<td>Previous MI, n/patient</td>
<td>1.8±0.6</td>
<td>1.5±0.6</td>
<td>0.2</td>
</tr>
<tr>
<td>Previous UA, n/patient</td>
<td>1.2±1.4</td>
<td>0.6±0.5</td>
<td>0.1</td>
</tr>
<tr>
<td>Stable angina, n (%)</td>
<td>4 (22)</td>
<td>6 (35)</td>
<td>0.5</td>
</tr>
<tr>
<td>Previous CABG, n (%)</td>
<td>8 (44.4)</td>
<td>5 (29.4)</td>
<td>0.5</td>
</tr>
<tr>
<td>Previous PCI, n (%)</td>
<td>13 (72.2)</td>
<td>12 (70.6)</td>
<td>1.0</td>
</tr>
<tr>
<td>Current smoker, n (%)</td>
<td>6 (33.3)</td>
<td>8 (47.1)</td>
<td>0.5</td>
</tr>
<tr>
<td>Ex-smoker, n (%)</td>
<td>10 (55.5)</td>
<td>9 (52.9)</td>
<td>1.0</td>
</tr>
<tr>
<td>Body mass index, kg/m(^2)</td>
<td>32.8±1.3</td>
<td>31.5±1.1</td>
<td>0.5</td>
</tr>
<tr>
<td>Waist circumference, cm</td>
<td>106.4±3.2</td>
<td>101.1±2.9</td>
<td>0.2</td>
</tr>
<tr>
<td>LDL-C, mmol/L</td>
<td>2.5±0.2</td>
<td>2.5±0.2</td>
<td>0.9</td>
</tr>
<tr>
<td>( \beta )-Blocker, n (%)</td>
<td>14 (77.8)</td>
<td>12 (70.6)</td>
<td>0.7</td>
</tr>
<tr>
<td>ACE inhibitor, n (%)</td>
<td>5 (27.8)</td>
<td>8 (47.1)</td>
<td>0.3</td>
</tr>
<tr>
<td>Calcium antagonist, n (%)</td>
<td>6 (33.3)</td>
<td>6 (35.3)</td>
<td>1.0</td>
</tr>
<tr>
<td>Long-acting nitrate, n (%)</td>
<td>5 (27.8)</td>
<td>2 (11.8)</td>
<td>0.4</td>
</tr>
<tr>
<td>Lipid-lowering agent (91% statins), n (%)</td>
<td>17 (94.4)</td>
<td>17 (100)</td>
<td>1.0</td>
</tr>
</tbody>
</table>

MI denotes myocardial infarction; UA, unstable angina; CABG, coronary artery bypass graft surgery; PCI, percutaneous coronary intervention; LDL-C, LDL cholesterol; and ACE, angiotensin-converting enzyme.
baseline values. Values of these markers at baseline and at 6 months of treatment, the primary object of analysis, are illustrated in Figure 1. The directional trends during the study of both CRP and IL-6 appeared similar. At 6 months, there was a significant lowering effect of rofecoxib compared with placebo on CRP ($P = 0.03$) and IL-6 ($P = 0.0002$). There was a significant off-drug effect in CRP and IL-6 levels in the rofecoxib group 3 months after the end of the treatment period ($P = 0.005$ and $P = 0.009$, respectively). There was a significant drug effect on P-selectin and MMP-9, the values of which remained relatively stable throughout the sampling period (Table 2).

There was a strong correlation between CRP and IL-6 values in the rofecoxib group ($r = 0.72; P < 0.0001$) but none in the placebo group ($r = 0.15; P = 0.3$). There was no significant drug effect on P-selectin and MMP-9, the values of which remained relatively stable throughout the sampling period (Table 2). Significant moderate correlations among the 4 markers and mediators that were evaluated (all values) were found between CRP and IL-6 ($r = 0.46; P < 0.0001$) and between P-selectin and MMP-9 ($r = 0.33; P = 0.0001$).

FMV and response to nitroglycerin at baseline and at 6 months in the rofecoxib and placebo groups are shown in Figure 2. One subject in the placebo group and 2 subjects in the rofecoxib group could not be analyzed because of inadequate echogenicity. There were no significant differences between the placebo and rofecoxib groups in FMV and NMV. The power of the ANOVA was estimated at 0.053; thus, there was a 94.7% chance of having committed a type II error. Therefore, these negative results would require a study of 1000 subjects to avoid missing a treatment effect on endothelial function that, if it existed, would likely be very small ($\approx 1\%$ lower FMV). Subjects with low baseline FMV in the rofecoxib group did not respond differently at 6 months compared with the placebo group. There were no correlations of serum values of CRP and IL-6 with either FMV or NMV.

**Discussion**

To the best of our knowledge, this is the first study to examine the effect of prolonged antiinflammatory treatment with a COX-2 antagonist on inflammatory markers and mediators and on vasoreactivity in subjects with ischemic heart disease. Additionally, in contrast to previous studies, these subjects were at distinctly high risk because of a history of recurrent acute coronary events and reproducibly high CRP levels.

The principal finding was a significant lowering effect of COX-2 inhibition on CRP and IL-6 compared with placebo. The pattern of response suggested an attenuating effect of

**TABLE 2. Values of CRP, IL-6, P-Selectin, and MMP-9 at All Time Points**

<table>
<thead>
<tr>
<th></th>
<th>Baseline</th>
<th>M1</th>
<th>M3</th>
<th>M6</th>
<th>PM1</th>
<th>PM3</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CRP, mg/L</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>3.16 (1.98, 5.52)</td>
<td>2.44 (1.85, 6.56)</td>
<td>2.57 (2.23, 4.72)</td>
<td>4.22 (2.32, 6.13)</td>
<td>2.50 (1.90, 4.06)</td>
<td>2.35 (1.59, 4.71)</td>
</tr>
<tr>
<td>Rofecoxib</td>
<td>3.45 (2.14, 5.52)</td>
<td>2.18 (1.81, 4.27)</td>
<td>2.90 (1.52, 5.07)</td>
<td>1.41 (1.18, 4.46)</td>
<td>2.84 (1.95, 4.23)</td>
<td>2.30 (1.58, 4.96)</td>
</tr>
<tr>
<td><strong>IL-6, pg/mL</strong></td>
<td></td>
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<tr>
<td>Placebo</td>
<td>2.7 (1.5, 3.1)</td>
<td>3.4 (2.6, 4.7)</td>
<td>2.8 (1.8, 3.6)</td>
<td>3.5 (1.9, 5.2)</td>
<td>2.4 (2.0, 3.0)</td>
<td>2.8 (2.1, 5.5)</td>
</tr>
<tr>
<td>Rofecoxib</td>
<td>2.3 (2.1, 3.4)</td>
<td>2.3 (2.0, 3.0)</td>
<td>2.6 (1.8, 3.4)</td>
<td>1.7 (1.4, 2.2)</td>
<td>2.1 (1.7, 3.6)</td>
<td>2.4 (1.9, 3.2)</td>
</tr>
<tr>
<td><strong>P-selectin, ng/mL</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>102.5±7.1</td>
<td>100.2±7.1</td>
<td>105.3±7.9</td>
<td>96.8±8.3</td>
<td>112.0±7.1</td>
<td>107.1±7.2</td>
</tr>
<tr>
<td>Rofecoxib</td>
<td>97.2±7.5</td>
<td>87.2±7.5</td>
<td>94.5±10.0</td>
<td>97.4±8.6</td>
<td>94.7±9.3</td>
<td>91.3±9.2</td>
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<tr>
<td><strong>MMP-9, ng/mL</strong></td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Placebo</td>
<td>326±41</td>
<td>351±56</td>
<td>353±47</td>
<td>315±60</td>
<td>319±45</td>
<td>309±52</td>
</tr>
<tr>
<td>Rofecoxib</td>
<td>359±44</td>
<td>304±34</td>
<td>329±34</td>
<td>303±40</td>
<td>303±36</td>
<td>275±48</td>
</tr>
</tbody>
</table>

M1, M3, and M6 indicate blood sampling at 1, 3, and 6 months of treatment, respectively; PM1 and PM3, blood sampling 1 and 3 months after study drug cessation. Values of CRP and IL-6 are median (25%, 75% quartiles); values of P-selectin and MMP-9 are mean±SEM.

* $P = 0.03$ vs baseline; † $P = 0.009$ vs M6; ‡ $P = 0.0002$ vs baseline; § $P = 0.005$ vs M6.
antiinflammatory therapy on CRP and IL-6 at 6 months, with a significant off-drug effect 3 months after treatment cessation. Values of CRP and IL-6 in the rofecoxib group were strongly correlated in contrast to the absence of correlation in the placebo group. Because CRP expression may be regulated by different mechanisms in different cells,29 this apparent rofecoxib-induced synchronization of CRP with IL-6 would be consistent with COX-2 inhibition of other inflammatory stimuli besides IL-6 on CRP expression. Serum P-selectin and MMP-9 appeared unaffected by rofecoxib. The latter is noteworthy because previous work showed a link within symptomatic atherosclerotic plaques between MMP-9 production and COX-2 activity and consequently a possible favorable effect of COX-2 inhibition on plaque stabilization.20 Our results suggest that blood levels of MMP-9 are not dependent on COX-2 activity at usual pharmacological doses.

We found no significant effect of prolonged COX-2 inhibition on endothelium-dependent brachial artery vasoreactivity. Previous studies suggested that antiinflammatory treatment has a neutral25,30 or favorable effect on endothelial function.31–33 On the other hand, CRP has recently been found to possess endothelium-independent vasodilating properties,34 so if CRP were reduced by COX-2 inhibition, a negative effect on vasoreactivity might have been noted. In addition, COX inhibition blunts prostacyclin production, and this might also have favored relative vasoconstriction. Complex opposing mechanisms could thus explain the absence of any apparent effect of COX-2 inhibition on brachial artery vasoreactivity. It may also be pertinent that baseline FMV in our subjects with coronary artery disease and enhanced inflammatory status was already in or close to the range found in normal subjects.35 Because study subjects had a favorable low-density lipoprotein cholesterol profile and nearly all were on chronic statin therapy, which has been shown to improve endothelial function,36 it may be that any additional vasodilation that COX-2 inhibition could exert on endothelial function had already been achieved.

Our findings extend and are also somewhat at variance to those of Chenevard et al,24 who, in a placebo-controlled crossover study of COX-2 inhibition with celecoxib in 14 subjects, found a small but significant improvement in endothelial function (increase in FMV from 2.0±0.5% to 3.3±0.4%) at 2 weeks with celecoxib and a significant lowering effect on serum CRP. In contrast, a recent study by Title et al25 in 60 subjects with stable coronary artery disease found no significant effect of rofecoxib, compared with placebo, on endothelial function, CRP, and soluble IL-6 receptor at 8 weeks compared with baseline. Differences in study design and drug and patient characteristics may account for these discrepancies. In contrast to these studies, which measured inflammatory markers at only 2 time points, our study was longer term with multiple measurements, including a prolonged off-drug period. Importantly, our recruitment criteria required consistently higher qualifying CRP values (2 of 2 values >2.0 mg/L) than the baseline values of the latter 2 studies. In fact, the baseline median CRP values in our rofecoxib and placebo groups were double those of the last 2 studies. Thus, the patients in our study may have constituted a more propitious group in which to test the effect of prolonged COX-2 inhibition.

These results must be tempered by the fluctuation of serum values of CRP and IL-6 that was manifest during screening, over the study period in the placebo group, and before 6 months in the rofecoxib group. This apparently spontaneous variability has been previously noted17,38 but is insufficiently appreciated because clinical studies have rarely examined serial values. Such variability might contribute to the discordant results of previous studies.24,25 Although our findings cannot be definitive, they do suggest that COX-2 inhibition attenuates the enhanced inflammatory profile of patients with previous recurrent acute coronary events.

Although the clinical literature on the cardiovascular effects of COX inhibitors is controversial1–7 our results are reassuring insofar as they suggest no deleterious effect—and possibly, a favorable effect—of such therapy on the inflammatory profile and no deleterious effect on vasoreactivity in a high-risk subset of patients with ischemic heart disease. Together with increasing recognition of the role of inflammatory markers and mediators in the atherosclerotic disease process and findings of recent basic14–16,20 and clinical24,25,39 studies, our results strengthen the rationale for exploring whether COX-2 inhibition may be beneficial in patients with ischemic heart disease who are at very high risk of acute coronary events because of their heightened inflammatory profile.51

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Impact of Prolonged Cyclooxygenase-2 Inhibition on Inflammatory Markers and Endothelial Function in Patients With Ischemic Heart Disease and Raised C-Reactive Protein: A Randomized Placebo-Controlled Study

Peter Bogaty, James M. Brophy, Martin Noel, Luce Boyer, Serge Simard, Fernand Bertrand and Gilles R. Dagenais

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