Clinical Investigation and Reports

Inflammation as a Possible Link Between Coronary and Carotid Plaque Instability

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Background—Multiple complex stenoses, plaque fissures, and widespread coronary inflammation are common in acute coronary syndromes. A systemic cause of atherosclerotic plaque instability is also suggested by studies of ischemic cerebrovascular disease. We investigated the association between coronary and carotid plaque instability and the potential common causal role of inflammation.

Methods and Results—The ultrasound characteristics of carotid plaques were evaluated retrospectively in patients scheduled for coronary bypass surgery, 181 with unstable and 92 with stable angina, and prospectively in a similar group of patients, 67 with unstable and 25 with stable angina, in whom serum C-reactive protein levels were also measured. The prevalence of carotid plaques was similar in the retrospective and prospective studies and >64% in both unstable and stable coronary patients. The prevalence of complex, presumably unstable carotid plaques was 23.2% in unstable versus 3.2% in stable patients (P<0.001) in the retrospective study and 41.8% versus 8.0% (P=0.002) in the prospective study. C-reactive protein levels were higher in patients with complex (7.55 mg/L) than in those with simple (3.94 mg/L; P<0.05) plaques or without plaques (2.45 mg/L; P<0.05). On multivariate analysis, unstable angina and C-reactive protein levels >3 mg/L were independently associated with complex carotid plaques (OR, 6.09; 95% CI, 1.01 to 33.72; P=0.039, and OR, 5.80; 95% CI, 1.55 to 21.69; P=0.009, respectively).

Conclusions—In unstable angina, plaque instability may not be confined to coronary arteries, and inflammation may be the common link with carotid plaque instability. These observations may have relevant implications for understanding the mechanisms of acute widespread atherothrombotic plaque inflammation. (Circulation. 2004;109:3158-3163.)

Key Words: angina ▪ carotid arteries ▪ inflammation ▪ plaque

Recent studies in patients with acute coronary syndromes show the presence of multiple complex coronary stenoses and fissures, consistent with reports of widespread inflammatory coronary plaque activation. Atherosclerotic plaque instability may not be confined to the coronary arteries but may also involve other arterial districts. This possibility is suggested by recent observations. First, in the European Carotid Surgery Trial, among patients with cerebrovascular accidents, those with angiographically irregular carotid plaques were more likely to have both irregular plaques in the contralateral nonculprit carotid artery and subsequent acute coronary events compared with patients having smooth carotid plaques. Second, the French Aortic Plaque in Stroke Group showed that complex aortic plaques, visualized by transesophageal echocardiography, were associated with a higher incidence of coronary events during follow-up than simple aortic plaques. Finally, in a small group of patients, complex carotid plaques were found to be associated with complex coronary stenoses. Infectious cell infiltrates were detected in multiple coronary plaques of patients who died of acute coronary syndromes and in carotid plaques of patients with recent cerebrovascular ischemic events. A common inflammatory link between acute coronary and carotid plaque instability would be consistent with the predictive value of C-reactive protein serum levels for myocardial infarction and for stroke in healthy men and women.

Therefore, we investigated whether (1) patients with unstable angina were more likely to harbor complex atherosclerotic plaques in the carotid arteries than patients with stable angina and (2) complex carotid plaques in these patients were associated with evidence of systemic inflammation. We addressed these questions in 2 consecutive studies. In a retrospective study, we evaluated the ultrasound morphological features of atherosclerotic plaques of carotid arteries in patients with unstable or with chronic stable angina scheduled for coronary artery bypass surgery. Then, we prospectively recruited a similar group of patients to assess the consistency of the findings of the retrospective study and to investigate...
the correlation between the prevalence of complex carotid plaques and serum levels of C-reactive protein.

Methods

Patients

In the retrospective study, we reviewed 223 consecutive patients admitted to our institute with Braunwald class IIB (42 patients) or IIIB (139 patients) unstable angina or with stable angina pectoris (92 patients), all of whom were submitted to B-mode ultrasound study of the carotid arteries as a part of routine diagnostic screening before coronary artery bypass surgery.

In the prospective study, we enrolled 92 consecutive patients admitted to our institute with Braunwald class IIB unstable angina (67 patients) or with chronic stable angina (25 patients) scheduled for either coronary artery bypass surgery or percutaneous coronary artery angioplasty.

Exclusion criteria were similar for both the retrospective and prospective studies: previous carotid endarterectomy/angioplasty, recent (<6 months) cerebrovascular accidents, and previous myocardial infarction. In the prospective study, patients with intercurrent inflammatory, infectious, or neoplastic conditions likely to be associated with an acute-phase response were also excluded. Unstable angina was defined as typical chest pain at rest or new-onset angina (in the previous 2 months) associated with diagnostic ECG ST-segment and/or T-wave changes without a diagnostic elevation of creatine kinase. Stable angina was defined as effort-related angina without any change in the clinical pattern in the preceding 2 months. From each patient, the following clinical data were obtained: age, sex, hypercholesterolemia (total plasma cholesterol concentration >200 mg/dL or ongoing lipid-lowering drug therapy), hypertension (any history of elevated blood pressure requiring antihypertensive therapy or a systolic blood pressure ≥140 mm Hg and/or a diastolic blood pressure ≥90 mm Hg in >2 measurements in hospital), diabetes mellitus (any history of diabetes or fasting plasma glucose >126 mg/dL or 200 mg/dL 2 hours after a meal), active smoking (≥5 cigarettes daily), and overweight/obesity (body mass index >25 kg/m²).

A subgroup of 54 patients with unstable angina of the prospective study underwent coronary angiography during the same hospitalization of the carotid ultrasonographic assessment. Coronary angiograms were reviewed by an expert interventional cardiologist (F.B.) who was unaware of clinical, laboratory, and ultrasonographic data. Significant (≥50%) coronary lesions were individuated and classified as simple or complex according to Ambrose classification. Serial or diffuse complex lesions were defined as multiple.1,2

The local ethics committee approved the protocol. All patients enrolled in the prospective study gave their informed consent to the use of some of their blood for the study.

Design and Procedures

Assessment of Carotid Arteries

Extracranial (common, internal, and external) carotid arteries were examined bilaterally in all patients with high-resolution B-mode and color Doppler/pulsed-wave Doppler ultrasonography (Toshiba SSA-270 or SSA-370 imaging system, 7.5/10-MHz linear-array transducer). All data of the retrospective study were collected and interpreted by 2 experienced ultrasonographers (A.L. and S.C.), and results were entered into an institutional database. In the prospective study, ultrasound images were stored on U-mathic videotape, and each examination was analyzed by the same 2 ultrasonographers blinded to any clinical information and laboratory results. Discrepancies were resolved by consensus. In case of persistent disagreement, a third expert reader (L.M.B.) was consulted, and a final decision was reached by consensus.

Carotid plaques were defined as a focal widening of the vessel wall relative to adjacent wall, protruding into the lumen. Plaques were analyzed for their extension, echogenic composition, and surface characteristics. Accordingly, carotid plaques were classified as complex when they had irregular surface and/or a heterogeneous echogenicity involving >50% of the plaque area and when they were mobile and/or ulcerated (Figure 1A); plaques were classified as simple when they were characterized by a smooth surface and a homogeneous hyperechoic structure involving >50% of the plaque area (Figure 1B).14-16 Patients who had both complex and simple carotid plaques were classified in the group of complex plaques. Carotid plaques were considered to cause a significant (≥50%) stenosis when the peak systolic Doppler flow velocity at the sites of maximal flow disturbance was >1.4 m/s.17

Measurement of C-Reactive Protein

In patients enrolled in the prospective study, venous blood samples to assess C-reactive protein levels were taken on admission and stored at −80°C until assayed. High-sensitivity C-reactive protein was measured in the serum with a commercially available high-sensitivity nephelometric method (Latex/BN II, Dade Behring). The working range of the assay was 0.175 to 1100 mg/L, and the coefficient of variation was <5%. All measurements were performed in a single batch at the end of the study by laboratory staff unaware of the clinical data. A cutoff value of high-sensitivity C-reactive protein of 3 mg/L was used to identify patients with elevated C-reactive protein levels.19

Statistical Analysis

Continuous variables were compared by unpaired t test, except for C-reactive protein levels, which were nonnormally distributed and were compared by the nonparametric Mann-Whitney U test. Discrete variables were compared by chi² test with Yates' correction. The prevalence of complex and simple carotid plaques and of normal (plaqueless) carotid arteries was analyzed according to high-sensitivity C-reactive protein tertiles. Univariate logistic regression analysis was used to assess the association of clinical and laboratory variables with the presence of complex carotid plaques. Multivariate logistic regression analysis was then applied to individuate the variables independently associated with the presence of complex plaque morphology. Only variables with a value of P≤0.1 on univariate analysis were included in the multivariate model. Age was entered as a continuous variable in regression analyses. Data are presented as mean±SD unless indicated otherwise. A value of P<0.05 was considered statistically significant. Analyses were performed with the statistical software SPSS 10.1 for Windows.

Results

Retrospective Study

The principal clinical characteristics of the patients included in the retrospective study are listed in Table 1. Overall, 187 (68.5%) of 273 patients were found to have atherosclerotic plaques in carotid arteries. No significant difference was found in the prevalence of carotid plaques in patients with unstable or stable angina (70.2% versus 65.2%, respectively; P=0.40). Similarly, no difference was found in the prevalence of carotid artery stenoses between the 2 groups (17.1% versus 13.0%, respectively; P=0.38).

However, complex carotid plaques were observed in 42 patients with unstable angina (23.2%) but in only 3 patients with stable angina (3.3%; P<0.001), whereas simple carotid plaques were observed more frequently in patients with stable angina than in those with unstable angina (61.9% versus 47.0%; P=0.019; Figure 2).

Univariate analysis revealed that unstable angina was the strongest predictor of complex carotid plaques (OR, 8.96; 95% CI, 2.7 to 29.8; P<0.001), followed by active smoking (OR, 2.85; 95% CI, 1.39 to 5.86; P=0.003) and age (OR, 1.06; 95% CI, 1.02 to 1.10; P=0.003; Table 2). On multivariate analysis, unstable angina (OR, 8.85; 95% CI, 2.59 to
30.28; P=0.001), active smoking (OR, 5.79; 95% CI, 2.39 to 13.99; P<0.001), and age (OR, 1.09; 95% CI, 1.04 to 1.11; P<0.001) were all independently associated with complex carotid plaque morphology.

**Prospective Study**
The clinical characteristics and cardiovascular risk factors of the 92 patients included in the prospective study are listed in Table 1. Overall, 68 patients (73.9%) were found to have atherosclerotic plaques of carotid arteries. There was no significant difference in the prevalence of carotid plaques in patients with unstable or stable angina (77.6% versus 64.0%, respectively; P=0.19). Similarly, there was no difference in the prevalence of carotid artery stenoses between the 2 groups (29.9% versus 20.0%, respectively; P=0.35).

**Figure 1.** Ultrasound images of complex and simple carotid plaques. A, Complex plaque of carotid artery bifurcation. Strongly heterogeneous echogenic structure is evident within plaque (left); irregular surface showing a crater suggestive of ulceration (arrow) is also discernible. Color flow at level of plaque is imaged (right). B, Simple plaque of common carotid artery (CCA). Homogeneous echogenic structure of plaque showing a smooth surface is evident (left). Normal color flow at level of plaque is also represented (right). ICA indicates internal carotid artery.

**TABLE 1. Clinical Characteristics and Cardiovascular Risk Factors of the 273 Retrospective and 92 Prospective Study Participants**

<table>
<thead>
<tr>
<th></th>
<th>Retrospective Study</th>
<th>Prospective Study</th>
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<tbody>
<tr>
<td></td>
<td>Unstable Patients</td>
<td>Stable Patients</td>
<td>P</td>
</tr>
<tr>
<td>Mean age, y</td>
<td>66.7±9.4</td>
<td>65.0±8.8</td>
<td>0.14</td>
</tr>
<tr>
<td>Men, n (%)</td>
<td>140 (77)</td>
<td>72 (78)</td>
<td>0.86</td>
</tr>
<tr>
<td>Hypercholesterolemia, n (%)</td>
<td>94 (52)</td>
<td>46 (50)</td>
<td>0.76</td>
</tr>
<tr>
<td>Hypertension, n (%)</td>
<td>111 (61)</td>
<td>55 (60)</td>
<td>0.81</td>
</tr>
<tr>
<td>Diabetes, n (%)</td>
<td>51 (28)</td>
<td>28 (30)</td>
<td>0.70</td>
</tr>
<tr>
<td>Active smoking, n (%)</td>
<td>32 (18)</td>
<td>17 (19)</td>
<td>0.87</td>
</tr>
<tr>
<td>Overweight/obesity, n (%)</td>
<td>70 (39)</td>
<td>34 (37)</td>
<td>0.78</td>
</tr>
<tr>
<td>Multivessel CAD, n (%)</td>
<td>153 (85)</td>
<td>71 (77)</td>
<td>0.13</td>
</tr>
<tr>
<td>Median hs-CRP (range), mg/L</td>
<td>...</td>
<td>...</td>
<td>...</td>
</tr>
<tr>
<td>Hs-CRP levels ≥3 mg/L, n (%)</td>
<td>...</td>
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</tbody>
</table>

CAD indicates coronary artery disease; hs-CRP, high-sensitivity C-reactive protein.
In the prospective study also, complex carotid plaques were more common in unstable than in stable angina (41.8% versus 8.0%; \( P = 0.002 \)). Conversely, simple carotid plaques were more common in patients with stable angina than in those with unstable angina, although this difference did not achieve statistical significance (56.0% versus 35.8%; \( P = 0.08 \)); Figure 2).

Serum C-reactive protein levels were significantly higher in patients with complex carotid plaques (median, 7.55 mg/L; range, 0.9 to 60 mg/L) compared with patients with simple carotid plaques (median, 3.94 mg/L; range, 0.5 to 26.6 mg/L; \( P < 0.05 \)) or without carotid plaques (median, 2.45 mg/L; range, 0.2 to 26.1 mg/L; \( P < 0.05 \)). Complex carotid plaques were detected more frequently in patients with high serum C-reactive protein (>3 mg/L) compared with patients with lower levels (44.6% versus 13.9%; \( P = 0.004 \)).

In univariate analysis, C-reactive protein serum levels >3 mg/L were significantly associated with complex carotid plaques (OR, 5.80; 95% CI, 1.55 to 21.69; \( P = 0.009 \)). Other univariate predictors of complex carotid plaques included diagnosis of unstable angina, active smoking, and age (Table 2). Multivariate logistic regression showed that active smoking (OR, 13.51; 95% CI, 2.94 to 67.46; \( P < 0.001 \)), C-reactive protein serum levels >3 mg/L (OR, 5.80; 95% CI, 1.55 to 21.69; \( P = 0.009 \)), unstable angina (OR, 6.09; 95% CI, 1.01 to 33.72; \( P = 0.039 \)), and age (OR, 1.14; 95% CI, 1.05 to 1.24; \( P = 0.001 \)) were independently associated with complex carotid plaques.

The prevalence of complex plaques increased significantly from the first to the third tertile of C-reactive protein concentrations, whereas the prevalence of simple plaques and of normal carotid arteries decreased (Figure 3).

In the subgroup of 54 patients who underwent coronary angiography, those with carotid plaques exhibited a higher number of diseased coronary vessels compared with those without carotid plaques (2.4 versus 1.8; \( P = 0.036 \)). Complex morphology was observed in 26% of the coronary stenosis found in patients without carotid plaques, in 34% of the coronary stenosis of patients with simple carotid plaques, and in 41% of the coronary stenosis of patients with complex carotid plaques (OR, 1.9; 95% CI, 0.9 to 4.4; \( P = 0.11 \), complex carotid plaques versus no carotid plaques). Moreover, multiple complex coronary plaques were found in 47.1% of patients with complex carotid plaques and in 21.6% of the patients without complex carotid plaques (OR, 3.2; 95% CI, 0.9 to 11.1; \( P = 0.057 \)).

### TABLE 2. Predictors of Complex Morphology of Carotid Plaques in the Retrospective and Prospective Studies at Univariate Analysis

<table>
<thead>
<tr>
<th>Variables</th>
<th>Retrospective Study</th>
<th>Prospective Study</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age</td>
<td>1.06 (1.02–1.10)</td>
<td>1.09 (1.03–1.16)</td>
</tr>
<tr>
<td>Male sex</td>
<td>0.75 (0.36–1.57)</td>
<td>0.80 (0.29–2.19)</td>
</tr>
<tr>
<td>Hypercholesterolemia</td>
<td>1.10 (0.58–2.09)</td>
<td>0.22 (0.03–1.94)</td>
</tr>
<tr>
<td>Hypertension</td>
<td>1.35 (0.69–2.65)</td>
<td>1.22 (0.46–3.21)</td>
</tr>
<tr>
<td>Diabetes</td>
<td>1.82 (0.94–3.55)</td>
<td>1.48 (0.61–3.59)</td>
</tr>
<tr>
<td>Active smoking</td>
<td>2.85 (1.39–5.86)</td>
<td>3.55 (1.34–9.38)</td>
</tr>
<tr>
<td>Overweight/obesity</td>
<td>0.98 (0.51–1.90)</td>
<td>0.63 (0.25–1.61)</td>
</tr>
<tr>
<td>Unstable angina</td>
<td>8.96 (2.7–29.8)</td>
<td>8.26 (1.80–37.91)</td>
</tr>
<tr>
<td>hs-CRP &gt;3 mg/L</td>
<td>...</td>
<td>5.00 (1.70–14.74)</td>
</tr>
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</table>

hs-CRP indicates high-sensitivity C-reactive protein.
Discussion

To the best of our knowledge, this is the first study to show that complex atherosclerotic plaques in carotid arteries are much more common in patients with unstable angina than in those with chronic stable angina. This indicates that in some patients with unstable angina, plaque instability may also involve the carotid arteries in a panvascular plaque activation, as suggested by a recent editorial, independently of the prevalence of conventional risk factor profile (Table 1). In the European Carotid Surgery Trial, ≈50% of patients with ischemic stroke exhibited irregular unstable plaques at angiography in the contralateral nonculprit carotid artery, and they were more likely to have fatal and nonfatal cardiac events than patients with smooth carotid plaques. The possibility of a common inflammatory link for such panvascular plaque activation is supported in our study by the independent correlation of high serum C-reactive protein levels with the presence of complex carotid plaques.

Ultrasound can provide detailed information on carotid plaque structure. In our study, carotid plaques were classified as complex or simple, depending on their echogenic features and surface morphology. Histological evidence suggests that homogeneous echogenicity is found in collagen-rich, fibrous plaques, and heterogeneous echogenicity is found in lipid-rich or hemorrhagic plaques, usually associated with an increased number of macrophages. The value of such classification is supported by its prognostic significance, because echolucent (complex) carotid plaques were found to be associated not only with a 3-fold-higher risk of ipsilateral ischemic stroke but also with a 2-fold-higher risk of cardiac ischemic events than echochir (simple) carotid plaques. Thus, such “complex” plaques appear to be potentially “unstable.”

The findings of the retrospective and prospective studies appear remarkably consistent, although the prevalence of complex plaques was higher in the prospective study, possibly because it included only patients with more severe instability (Braunwald class IIIB). This novel information, provided by ultrasonography, could be investigated in greater detail with high-resolution MRI and PET, which can assess composition and inflammation in atherosclerotic plaques more accurately.

Multifocal Plaque Instability

The most obvious morphological feature of the plaque in acute coronary syndromes is represented by thrombosis at the site of a “culprit” coronary plaque. In addition, ischemic cerebrovascular accidents are believed to be most commonly caused by thromboembolic events originating at the site of a “culprit” carotid plaque. Several recent findings, however, suggest multifocal coronary plaque instability in patients with acute coronary syndromes, possibly as an expression of a widespread coronary inflammation involving also arteries without angiographically detectable stenoses. These clinical findings were confirmed by postmortem observation of multiple inflamed plaques in different coronary branches of patients who died as a result of acute coronary syndromes. Moreover, the simultaneous presence of multiple complex atherosclerotic plaques was also reported in arterial districts other than the coronary arteries.

Inflammation and Multifocal Plaque Instability

Acute coronary and cerebrovascular syndromes as well as severe peripheral artery disease are commonly associated with systemic evidence of inflammation. Elevated serum high sensitivity C-reactive protein levels were found in 50% to 70% of patients with each of these vascular syndromes and are associated with worse outcome. Moreover, high-sensitivity C-reactive protein levels predict the progression of atherosclerotic plaques in the arterial tree and its persistent elevation predicts recurrent instability.

A common link between coronary and carotid plaque instability may result indirectly from the activation of resident inflammatory cells in carotid plaques by circulating activated leukocytes and proinflammatory cytokines or alternatively from the same primary triggers specifically responsible for the localization of inflammation in coronary arteries. The possible existence of common inflammatory pathogenetic mechanisms in patients with acute coronary syndromes and in those with cerebrovascular ischemic events may, at
least in some patients, explain the findings of the European Carotid Surgical Trial\(^6\) and would be consistent with the 50% reduction in cerebrovascular events achieved with atorvastatin in the Myocardial Ischemia Reduction with Aggressive Cholesterol Lowering (MIRACL) trial during a 16-week follow-up after acute coronary syndromes.\(^{32}\)

However, the significant independent correlation of complex carotid plaques with smoking and with unstable angina observed in the multivariate analysis suggests that the relationship between plaque instability and C-reactive protein is a complex one.

**Conclusions**

Taken together, our findings suggest that patients with unstable angina, particularly those with elevated C-reactive protein, may have not only widespread coronary inflammation and multiple complex coronary plaques but also morphological characteristics of instability of carotid artery plaques that, in turn, might result in cerebrovascular ischemic events or in rapid plaque growth. Patients with unstable angina should undergo routine checks for complex carotid plaques. In such patients, the study of carotid plaque inflammation could provide useful insights into the local and systemic mechanisms of atherosclerotic plaque instability.

**Acknowledgments**

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**References**


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