Seeing and Sampling the Surface of the Atherosclerotic Plaque
Red or White Can Make Blue

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Understanding the mechanisms underlying acute coronary syndromes (ACS) depended for decades on autopsy studies. The limitations of postmortem studies include death as an inclusion criterion, biasing the studies toward events with fatal outcomes. The more numerous nonmortal events comprise a sizable but heretofore largely hidden denominator of uncertain magnitude. To gain insight into the underlying pathophysiologic basis of the ACS, laboratory researchers have conducted innumerable animal studies on atherosclerosis. Advanced genetic manipulations have led to increasingly refined approaches to probe the aspects of experimental atherosclerosis, particularly in mice. Despite their indisputable value for isolating mechanisms, the interpretation and extrapolation of the results of many such mouse studies blithely ignore the yawning gap between the disease in laboratory animals and human patients with atherosclerosis. Only under extreme circumstances do mouse or rabbit atheromata actually ever cause thrombosis. Experiments in small animals almost never focus on the coronary arteries, structures that differ strikingly in ontogeny from the human aorta or carotid arteries. Experimental studies routinely refer to plaque vulnerability or stable or unstable plaque phenotype without actually assessing stability, instability, or vulnerability. This leap of faith, albeit routinely accepted, represents a striking suspension of intellectual and scientific rigor to which many turn a blind eye.

Fortunately, we now possess tools of increasing utility and validation for probing the structure of atherosclerotic plaques in living humans. Traditional contrast arteriography provided silhouettes of the lumen but revealed little of the artery wall or the character of atherosclerotic plaques themselves. Intravascular ultrasound provides an outstanding modality for measuring the volume of atherosclerotic plaques. Use of the radiofrequency backscatter from intravascular ultrasound can furnish some rudimentary information about tissue characterization. With the advent of optical coherence tomography (OCT), we now possess a remarkable method for near-field imaging of the arterial intima in intact humans. OCT allows visualization of the surface of the plaque and the near-field subjacent intima with almost microscopic resolution. Although OCT does not furnish precise cellular or biochemical information, its practitioners have developed criteria for distinguishing rupture of the plaque’s fibrous cap from nonrupture, and red versus white thrombus. Either red or white thrombus can provoke myocardial ischemia (recognizable as cyanosis or blue discoloration of the heart’s surface). There is even evidence that OCT can identify macrophages in intimal lesions, although this feature may be ambiguous. The judicious application of this powerful imaging modality can provide novel insights about the correlation of mechanisms of plaque disruption, thrombosis, clinical outcomes, and biomarkers. This rich opportunity points to new ways to probe pathophysiologic pathways in humans and to permit more personalized, precision approaches to therapy.

Nishiguchi et al in this issue of Arteriosclerosis, Thrombosis, and Vascular Biology use OCT to these ends in a series of patients presenting with coronary artery disease. Appropriately, most of the patients requiring acute intervention had ST-segment–elevation myocardial infarction (STEMI). These patients underwent manual thrombus aspiration and subsequent sampling of the coronary arterial blood in the region of the culprit lesion before and after the deployment of a stent and in peripheral blood. The authors report on concentrations of 2 biomarkers measured in the samples: matrix metalloproteinase-9 (MMP-9) and myeloperoxidase. They found a significant elevation in the local versus systemic blood concentrations of MMP-9 in patients with STEMI, albeit with considerable overlap. They furthermore found more thrombi that display OCT signatures of red (presumably fibrin-rich) clots in STEMI than in patients who presented with non–ST-segment–elevation myocardial infarction.

These findings raise several interesting issues with relation to the study itself, its insights into understanding the pathophysiology of ACS and its implications for personalized therapy. To perform OCT in the safest manner in patients undergoing an ACS, the investigators performed a preliminary thrombus aspiration. Yet, one must ask whether this manipulation could itself alter the release or mobilization of the analytes tested. As in accord with the Heisenberg uncertainty principle in quantum physics, could the very act of gazing at the plaque so prepared change the measurements made? The choice of myeloperoxidase for investigation in this study derives from previous observations that
demonstrated that concentrations of this pro-oxidant enzyme rise to a greater extent in ACS caused by superficial erosion than by plaque rupture. As the authors point out, their patient population was biased toward STEMI, as appropriately, many non–ST-segment–elevation myocardial infarction patients did not undergo urgent intervention. That feature may explain, in part, why this study did not find an association between the type of presentation and local myeloperoxidase concentration. The choice of MMP-9 as a focus for this study is less clear. The finding of predominantly red thrombi in this STEMI-slanted population agrees with the notion that lesions that provoke ACS because of superficial erosion may have more platelet-rich, white thrombi. Was MMP-9 a prespecified target of this investigation? How many other biomarkers did these investigators analyze that might not have shown significant gradients between coronary arterial and systemic blood? Did the authors analyze MRP 8/14 that we and others have implicated as a biomarker and actual agonist in STEMI? 

Because MMP-9 did emerge as significantly elevated in the coronary arterial blood sampled at the site of the culprit lesion, the mechanistic implications of this observation merit consideration. Early studies localized this enzyme in human atherosclerotic plaques. This enzyme has many functions that could relate to atherogenesis and plaque disruption (Figure). As a potent elastase, MMP-9 could participate in smooth muscle migration and in outward (Glagovian) geometric remodeling of plaques during their evolution. As a type IV nonfibrillar collagenase, MMP-9 could degrade the intimal basement membrane, rendering endothelial cells more likely to detach. Sites with impaired or absent endothelial antithrombotic and fibrinolytic and vasodilator function might propagate or aggravate local thrombus accumulation. MMP-9 can cleave prointerleukin-1β to produce the mature, active form of this proinflammatory cytokine. As the authors point out, MMP-9 can also degrade tissue factor pathway inhibitor, removing a homeostatic brake on coagulation. MMP-9’s ability to affect such local changes in atheroma would require the presence of the enzyme’s active form rather than the inactive zymogen precursor proteinase (Figure). The assay for MMP-9 used in the study likely could not distinguish between the active and inactive forms of this enzyme, leaving this point unresolved.

As the authors argue, biomarkers that could stratify patients with ACS mechanistically and therapeutically might help achieve the goal of a more precision management and personalized approach. Although MMP-9 could serve as such a marker, several practical obstacles impede its implementation in clinical practice. There was a high degree of overlap in MMP-9 measurements between groups, rendering this analyte difficult to use as a binary biomarker to inform diagnosis and management. MMP-9 measurement in coronary arterial blood requires invasive instrumentation, whereas an ideal strategy for precision triage of patients with ACS would avoid catheterization in a subset of individuals who might be managed without interventional treatment. MMP-9’s measurement in the ACS setting would also require near instantaneous point-of-care testing, a potentially feasible undertaking, but one not yet in hand or validated. This study provides important illustration of the principle that biomarkers may serve to discriminate various substrates for ACS that could direct individuals to different management strategies. The concept is powerful, pertinent to precision and personalized medicine, and provides a worthy goal toward which to strive in the future.

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None.

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**Figure.** Selected actions of matrix metalloproteinase-9 (MMP-9) related to atherothrombosis. Pro-MMP-9 undergoes cleavage to generate the active MMP-9 catalytic domain. Active MMP-9 then can process prointerleukin-1β (IL-1β) to form the proinflammatory active cytokine. MMP-9 also exhibits elastolytic activity that can mediate arterial remodeling and smooth muscle cell migration. MMP-9 also possesses gelatinolytic activity that can degrade type IV collagen, a major constituent of the subendothelial intimal basement membrane. Catabolism of collagen IV may favor endothelial cell sloughing, potentially promoting local thrombus extension. MMP-9 can also inactivate tissue factor pathway inhibitor (TFPI). This action of MMP-9 could promote thrombosis by removing an endogenous inhibitor of the potent procoagulant tissue factor, implicated in coronary artery thrombus formation. The domains of promatrix metalloproteinase-9 depicted include the signal sequence (S), the Pro-piece (Pro-), the Collagen V-like domain (Col V), and the hemopexin (HP) domain at the carboxyl terminus of the zymogen.
References


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