Concept of Vulnerable/Unstable Plaque

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Abstract—Today’s concept of vulnerable plaque has evolved primarily from the early pioneering work uncovering the pivotal role of plaque rupture and coronary thrombosis as the major cause of acute myocardial infarction and sudden cardiac death. Since the first historical description of plaque rupture in 1844, several key studies by leading researchers and clinicians have lead to the current accepted views on lesion instability. Important to the complex paradigm of plaque destabilization and thrombosis are many discoveries beginning with the earliest descriptions of advanced plaques, reminiscent of abscesses encapsulated by fibrous tissue capable of rupture. It was not until the late 1980s that studies of remodeling provided keen insight into the growth of advanced plaques, beyond the simple accumulation of lipid. The emphasis in the next decade, however, was on a focused shift toward the mechanisms of lesion vulnerability based on the contribution of tissue proteolysis by matrix metalloproteinases as an essential factor responsible for thinning and rupture of the fibrous cap. In an attempt to unify the understanding of what constitutes a vulnerable plaque, morphological studies, mostly from autopsy, suggest the importance of necrotic core size, inflammation, and fibrous cap thickness. Definitive proof of the vulnerable plaque, however, remains elusive because animal or human data supporting a cause-and-effect relationship are lacking. Although emerging imagining technologies involving optical coherence tomography, high-resolution MRI, molecular biomarkers, and other techniques have far surpassed the limits of the early days of angiography, advancing the field will require establishing relevant translational animal models that produce vulnerable plaques at risk for rupture and further testing of these modalities in large prospective clinical trials. (Arterioscler Thromb Vasc Biol. 2010;30:1282-1292.)

Key Words: acute coronary syndromes ■ coronary artery disease ■ pathology ■ thrombosis ■ vulnerable plaque

Despite recent advances in medical and interventional percutaneous or surgical therapies, coronary artery disease continues to be a major cause of morbidity and mortality throughout the world. In the United States alone, coronary artery disease is responsible for a majority of deaths, accounting for more than 400,000 (≈1 of every 6 deaths) annually.1 The typical patient presents with acute coronary syndromes (ACS) defined by acute myocardial infarction (AMI) or unstable angina, where approximately 300,000 patients succumb to the initial coronary event. Modern cardiology is focused on developing techniques designed to restore blood flow in arteries possessing hemodynamically significant lesions causing cardiac ischemia and infarction. This reactive strategy, however, does little to prevent future coronary events. Although pharmacotherapy with agents such as aspirin and statins has been proven to lower the risk of future coronary events, it does not represent a panacea.

Major advances in coronary artery disease prevention will require early detection of rupture-prone or so-called vulnerable plaques. Therefore, it is essential to further identify meaningful surrogates of lesion instability at the highest risk of rupture. Moreover, vulnerable plaques tend to occur at multiple sites, and some have advocated the concept of the vulnerable patient, defined by high atherosclerotic burden, high-risk/vulnerable plaques, or thrombogenic blood. Paradoxically, these patients might be recognized by biomarkers other than those directed toward specifically identifying vulnerable plaques.

History of the Vulnerable Plaque

An understanding of the pathophysiology of vulnerable plaque would not have been possible without the pioneering work that came before the elucidation of the pivotal role of plaque rupture with superimposed coronary thrombosis as the major cause of myocardial infarction (MI) (Figure 1).

The concept of plaque rupture was first reported at the autopsy of the celebrated neoclassical Danish artist Bertel Thorvaldsen, who died of sudden cardiac death in the Royal Theater in Copenhagen in 1844.2,3 It was not until early in the next century that researchers focused on pathological features of culprit lesions responsible for sudden cardiac death. Notable investigators such as Clark, Koch, Friedman, and Constantinides were the first to describe fissures and erosions on the intimal surface of coronary arteries as the causation of thrombosis.4-6 Around the same time, Wartman and others, such as Patterson and Winternitz, noted the importance of

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intramural hemorrhage in the progression of coronary lesions.\textsuperscript{7–9} Using a systematic approach of serial sectioning, Friedman made significant inroads describing many of the findings that established today’s understanding of plaque rupture. Adopting descriptive terms such as “intramural atheromatous abscess” (first reported by Leary\textsuperscript{10}), Friedman demonstrated communication of necrotic material with the occluding thrombus in 39 of 40 arteries, concluding that rupture exposes the blood to highly thrombogenic contents of the plaque, leading to clot formation.\textsuperscript{11} Another pioneering pathologist, Michael J. Davies (1937–2002), devoted himself to the study of plaque rupture and its associated features, describing in detail the features of plaque disruption and the role of inflammation in the development of plaque instability.\textsuperscript{12,13} Landmark reviews from a myriad of notable investigators forwarding the more recent concepts of lesion vulnerability became available in the mid-1990s, in particular the classic article “Coronary Plaque Disruption” by Erling Falk et al.\textsuperscript{14}

As the understanding of the pathophysiology of lesion instability began to expand, there was a necessity for consensus terms regarding vulnerability; the adoption of a common nomenclature would help support the study of acute coronary events. To meet this need, a group of prominent investigators convened in 2003 to resolve issues of terminology surrounding the identification of high-risk and vulnerable coronary artery plaques.\textsuperscript{15} A unification of conceptual terms was adopted to provide a fundamental basis for clinicians and researchers whereby diagnostic methods of identifying vulnerable patients and the plaques that contribute to their increased risk can be identified, in addition to defining the critical lesions that may be amenable to treatment.

Although coronary thrombi were a frequent finding at autopsy, the causative nature of thrombosis in the pathogenesis of AMI was contested for decades. Several researchers emphasized the role of thrombosis in AMI; however, the 1960s and 1970s saw fierce debate over whether thrombus formation followed or preceded AMI, fueled in part by the inconsistent reporting of thrombi in fatal MIs. A negative proponent, the noted pathologist William C. Roberts, believed that “the virtual absence of coronary thrombi in patients dying suddenly of cardiac disease, and their presence in only about one-half of those with myocardial necrosis, supported the concept that coronary arterial thromboses were consequence rather than cause of AMI.”\textsuperscript{16}

It was not until 1980 that DeWood et al provided definitive angiographic evidence that intracoronary thrombi have a causal role in the pathogenesis of acute coronary occlusion in AMI.\textsuperscript{17} An earlier study by Fulton and Sumner, using premortem injection of \textsuperscript{125}I-labeled fibrinogen, gave rise to the notion that thrombosis often preceded MI.\textsuperscript{18} The greater understanding of the essential role of thrombosis in the causation of MI led to the introduction of thrombolysis as a means of treating infarction patients in the coronary care units. This preceded the now well-accepted “open artery” theory for all MI patients, where the current recommended
door-to-balloon time using primary percutaneous coronary intervention is \(\leq 90\) minutes.19

Advancement in our understanding of the pathophysiology of MI was an essential step in the definition of vulnerable plaque. Retrospective angiographic studies of patients presenting with AMI promoted the idea that MI frequently develops in previously nonsevere lesions.20 In 1989, James E. Muller et al. named these hemodynamically insignificant plaques vulnerable plaques, defined as lesions with a susceptibility to rupture.21

More recently, our laboratory and others have defined other etiologies of luminal thrombosis, namely plaque rupture, erosion, and calcified nodules.22 Plaque rupture refers to a lesion consisting of a necrotic core with an overlying thin ruptured fibrous cap that leads to luminal thrombosis because of contact of flowing blood with a highly thrombogenic necrotic core.23 On the other hand, plaque erosion shows a lesion with a thin cap, but when present, the core does not communicate with the lumen because of the thickened fibrous cap. The least common of all lesions giving rise to acute coronary thrombosis is the calcified nodule, recognized by calcified plates with superimposed calcified bony nodules that result in discontinuity of the fibrous cap, with an irregular luminal surface devoid of endothelial cells and overlying luminal thrombus.22

From our examination of more than 800 cases of sudden coronary death at autopsy, 55% to 60% of subjects have underlying plaque rupture as the etiology, whereas for 30% to 35% the etiology is erosion and for 2% to 7% it is thrombi attributed to calcified nodules. Plaque rupture is also the predominant cause of death at autopsy, occurring in 75% of patients presenting with AMI (mean age, men = 68 ± 11, women = 70 ± 9 years) diagnosed by ECG and enzyme elevation.25 In contrast, approximately 37% of women with AMI had plaque erosion, whereas in men erosion was present in only 18% (\(P = 0.0004\)).25 Overall, plaque erosion appears to be the primary cause of acute coronary thrombi in women under the age of 50 years who present with sudden coronary death.24 Thus, the etiology of the thrombus is dependent on age and sex, where plaque rupture is a dominant mechanism in men regardless of age and in older, postmenopausal women older than 50.26

From a pathologist’s perspective, the understanding of vulnerable plaque is mostly derived from comprehensive examinations of plaque rupture simply because of the failure to identify precursor lesions of erosions or the calcified nodule. The lack of other precursor lesions besides rupture represents a major shortcoming to the vulnerable plaque paradigm, as discussed below.

**Pathological Characterization of the Thin-Cap Fibroatheroma or Vulnerable Plaque**

The morphological criterion that supports the definition of thin-cap fibroatheroma (TCFA) or vulnerable plaque results from an astoundingly simple tenet that the preceding lesion on the road to rupture bears a strong resemblance in morphology to rupture itself (Figure 2). A necrotic core characterizes plaque rupture with an overlying thin-ruptured cap infiltrated by macrophages and lymphocytes. There are few or no smooth muscle cells within the cap. The thickness of the fibrous cap near the rupture site measures 23 ± 19 \(\mu\)m, with 95% of caps measuring <65 \(\mu\)m.27 It is precisely those lesions with intact fibrous caps of <65 \(\mu\)m observed at other (oftentimes multiple) sites in the coronary vasculature, in patients dying of acute plaque rupture, that are designated vulnerable plaques or TCFA. Therefore, the current definition of vulnerability is exclusive to plaque rupture.

Despite morphological similarities, TCFA differ from ruptures considering that they generally exhibit smaller necrotic cores, fewer macrophages within the fibrous cap, and less calcification.28,29 Although the vulnerable plaque was not a recognized entity in the American Heart Association consensus document, it was highlighted in our modified classification scheme published later in 2000.22 One of the major limitations of the original American Heart Association classification was a lack of understanding that the fibrous cap undergoes thinning before the onset of rupture, a concept forwarded by Dr Peter Libby30 (Figure 3) and further defined by our laboratory.27

**Major Turning Points Toward an Understanding of the Pathophysiology of Lesion Instability**

**Lesion Growth in Advanced Plaques Differs Significantly From Early Lesions**

It gradually became apparent that 2 key processes are involved in the growth of advanced plaques: (1) outward, abluminal expansion of the arterial wall and (2) subclinical plaque rupture of hemodynamically insignificant lesions, where the thrombus is incorporated into the lesion, resulting in greater luminal narrowing. Historically, these beliefs were slow to evolve because of the understanding of lesion progression in the clinic was, and still is, mostly based on angiography, which cannot determine plaque morphology. The prevailing notion surrounding lesion growth began to change, however, following publication of a seminal study by Sey mour Glagov et al in 1987, reporting that progressive plaques endure a lengthy phase of positive remodeling or arterial wall expansion (occurring over years or decades) that is responsible for the preservation of lumen size.31 Luminal compromise only occurs once the plaque advances beyond 40% narrowing, ie, when positive remodeling stops. Davies forwarded the concept that repeated silent plaque ruptures are the cause of luminal compromise, which heal and lead to severe luminal narrowing.32 Similarly, we reported in sudden death subjects that plaques progress through repeated ruptures (ie, luminal narrowing occurs through healed plaque ruptures) and that only 11% of acute plaque ruptures are virgin ruptures.33

**Rendering a Plaque Unstable: The Emergence of Inflammation and Tissue Proteolysis as an Underlying Mechanism of Plaque Vulnerability**

A seminal article by Russell Ross et al. described the presence of macrophages within the fibrous cap in lesions from the
superficial femoral artery collected at surgery. He noted that the majority of fibrous caps overlying lipid cores from lesions with superficial thrombi contained smooth muscle cells with varying numbers of macrophages. What makes a plaque stable is the intact and thick fibrous cap that is made up of smooth muscle cells in a matrix rich in type I and III collagen. Richardson and coworkers performed mechanical tests on strips of fibrous caps with infiltrating macrophages from human aortic samples with ulcerated or intact plaque caps and showed a good correlation of increase in macrophage density with greater extensibility and decreased maximum stress (force per unit area).

Key observations in the early to mid-1990s by Falk, Shah, and Moreno implicating the significance of proteolytic enzymes released from macrophages in lesions responsible for unstable angina, helped establish the paradigm of matrix degradation and lesion instability. Around the same time, Libby greatly expanded on this hypothesis, proposing that collagen synthesis within the fibrous cap was impaired and that matrix degrading enzymes, a main component of the vast secretory repertoire of the macrophage, may ultimately be responsible for degradation of the fibrous cap. Elegant in vitro studies revealed the importance of interferon-γ, which is secreted by activated T cell and is responsible for the markedly decreased ability of human smooth muscle cells to express interstitial collagen genes in both resting and stimulated states. T cells synthesize interferon-γ, and in the fibrous cap, they exhibit nonuniform patterns of type I collagen gene expression, which is negatively associated with the spatial presence of T cells.

The perception of increased matrix degradation as a mechanism of lesion instability was mainly based on the early
The loss of structural molecules provided by the extracellular matrix can thin and weaken the fibrous cap, rendering it particularly susceptible to rupture and acute coronary syndromes. Additional factors involved in the activation of macrophages include tumor necrosis factor-α (TNF-α), macrophage colony-stimulating factor (M-CSF), and macrophage chemoattractant protein-1 (MCP-1), among others. Importantly, the expression of CD40 ligand on T cells may promote tissue proteolysis through the release and activation of matrix-degrading enzymes produced by vascular smooth muscle cells and inflammatory macrophages. Activated macrophages within the fibrous cap can secrete tissue proteases that support the breakdown of collagen and elastin to peptides and amino acids. The studies published by Henney et al demonstrating the presence of stromelysin mainly in macrophages in coronary arteries. In the mid 1990s, Zorina Galis et al demonstrated 3 matrix metalloproteinase (MMP) classes (interstitial collagenase, MMP-1; gelatinases, MMP-2 and MMP-9; and stromelysin, MMP-3) expressed primarily in the shoulder regions of advanced plaques along with their endogenous inhibitors (tissue inhibitors of matrix metalloproteinases [TIMPs] 1 and 2). MMP enzymatic activities were demonstrated by in situ zymography (in vitro, lysis of a thin film of collagen substrate) showing focal overexpression of activated MMP as a potential mechanism that promotes destabilization. Furthermore, Shah et al showed that monocyte-derived macrophages exposed in vitro to fibrous caps dissected from human aortic or carotid plaques resulted in degradation of the fibrous cap.

**Incidence and Frequency of the TCFA**

Burke et al, from our laboratory, not only defined vulnerable plaques (as defined above) but also were among the first to show that vulnerable plaques were commonly observed in patients dying a sudden coronary death and were more common in plaque ruptures than in stable plaques. The number of vulnerable plaques correlated with both high total cholesterol and the total cholesterol/high-density lipoprotein cholesterol ratio. In subjects dying of plaque rupture, additional lesion sites remote from the culprit plaque typically show TCFAs in 70% of cases. On the other hand, TCFAs are less frequent (30%) in cases where death is attributed to fibrocalcific plaques with flow-limiting stenosis, regardless of MI status, or plaque erosion. Therefore, it must be emphasized that not all TCFAs are likely to progress to rupture; however, it is important to further define the essential surrogates of lesion instability at the highest risk of rupture.

In an additional 38 hearts from sudden coronary deaths with severe luminal narrowing, in which the arteries had been serially cut from coronary ostium to a distal intramyocardial location, mean luminal narrowing was least in sections with TCFAs (59.6%), intermediate for lesions with hemorrhage into a plaque (68.8%) and greatest in acute plaque ruptures (73.3%) or healed plaque ruptures (72.8%). Overall, nearly 75% of lesions showed <75% cross-sectional luminal-narrowing or (<50% diameter stenosis), which may be a useful indicator for the detection of vulnerable plaque. Moreover, the location is also important, as approximately 50% of the TCFAs occur in the proximal portions of the major coronary arteries (left anterior descending > left circumflex > right coronary artery), with another one third in the midportion and the remaining few in distal segments. A similar regional distribution of TCFAs is found for acute and healed plaque ruptures. Clinical studies in AMI patients also confirm that the proximal portions of all 3 major coronary arteries are the most common locations for thrombotic occlusion.

Remodeling, as mentioned above, can also be an important sign of vulnerability. In this regard, plaque ruptures had the highest remodeling index, followed by lesions with hemorrhage > TCFAs > healed plaque ruptures > fibroatheromas. Conversely, lesions of total occlusion or erosion exhibited negative remodeling. From this study, it becomes evident that all lesions derived from or related to plaque rupture show positive remodeling, which may represent one important surrogate for detecting lesion vulnerability.

**Mechanical Stress, Lesion Vulnerability, and Rupture**

A limited number of biomechanical and imaging studies started to emerge in the early 1990s addressing the role of hemodynamic shear stress in the destabilization of vulnerable plaques. The underlying premise is based on observations that atherosclerosis is a focal point, where rupture occurs more frequently at the proximal side of the stenosis.
near flow dividers, an area where secondary wall shear stress is assumed to be the highest. Consistent with these observations, blood flow–induced shear stress may also present a significant influence on processes that govern fibrous cap morphology and composition, where increased peak circumferential stress is greater in thinner fibrous caps. Therefore, regions of high shear stress typically exhibit high strain, thus supporting the notion that mechanical stress applied to a weakened fibrous cap may precipitate rupture, particularly in the presence of microcalcification.

**Limitations to the Vulnerable Plaque Paradigm**

From the view of vulnerable plaque detection, morphological studies should provide valuable insight regarding which criteria are important for localizing high-risk lesions using imaging modalities and assessing the potential risk for rupture. Obvious important cause-and-effect data are missing from the current paradigm because of our inability to accurately detect vulnerable plaques in humans in vivo and because of the lack of an animal model. In this regard, representative lesions in current animal models rarely progress beyond the stage of fibroatheroma; more often, they consist of only masses of lipid-laden intimal macrophages without a well-developed fibrous cap or necrosis. Lesions with this histology rarely become clinically significant except in examples of severe hyperlipidemia, in which the lumen can become obstructed by the shear plaque burden, a situation that is quite atypical in human disease. Therefore, a major limitation today exists partly because the precise mechanisms of progression from an asymptomatic stable to high-risk plaque (TCFA or vulnerable plaque) that lead to rupture and thrombosis are incompletely understood.

Although mouse models of atherosclerosis have provided important insights into the genetic component of the disease, major differences in the pathways directed toward lesion formation between mice and humans remain apparent. For example, inflammation is the most abundant plaque component in mice, whereas in humans it constitutes only 2% to 5% of total lesion volume. Moreover, intraplaque hemorrhage from leaky vasa vasorum in human plaques is believed to be a significant factor in necrotic core expansion, a phenomenon rarely observed in mice. Perhaps more importantly, the mechanisms of disease initiation and progression are quite distinct, considering that mouse lesions are products of macrophages, whereas in humans, early lesion progression arises from smooth muscle cells enriched in an environment of proteoglycans and collagen with discreet pools of lipid, so-called pathological intimal thickening. Although features of fibrous cap formation, protease, activation, and cell death in mice are reminiscent of human plaques, the applicability of these to the natural progression of the disease has not yet been tested. Therefore, we have proposed an alternative lesion classification for mice in which early to intermediate lesions are represented by varying degrees of macrophage infiltration. Rather than rupture per se, the pinnacle of advanced plaques in mice appears to be the activated atheroma, characterized by focal medial degradation and extensive adventitial inflammation, including mast cells with neovascularization.

**Current Status of Vascular Imaging as an Understanding Toward the Natural History of the Vulnerable Plaque**

To advance beyond the reactive clinical strategy that defines modern cardiology, significant advances in our understanding and natural history of TCFA (as well as other plaque morphologies) can only be achieved by improving our ability to accurately define and locate these lesions in the clinic. Detection based on morphology alone has been proposed using imaging modalities such as intravascular ultrasound. Although some articles have claimed to visualize ruptured plaques using this technology, it is doubtful, considering the limited resolution (150 to 200 μm) of this modality. Perhaps more usefully, ultrasonography has proven successful in the carotid circulation to predict the future risk of stroke based on echolucency. Histologically, echolucent plaques generally have higher lipid content, macrophage density, and hemorrhage.

Among the current available modalities, MRI emerges as most promising for assessing plaque morphology, in particular for assessing aortic and carotid plaques at risk for embolic stroke. Its superior capability of determining plaque size and composition with reasonable accuracy and reproducibility provides opportunities to study relationships between plaque morphology and composition and subsequent cerebrovascular events. In this regard, Takaya et al tested the hypothesis that the characteristics of carotid plaques assessed by MRI are future predictors of ipsilateral cerebrovascular events in 154 asymptomatic subjects with at least 12-month follow-up with definitive stenosis (50% to 70%) by ultrasound. Correlates of lesion structure and composition with associated cerebrovascular events included thinned or ruptured fibrous caps, presence of intraplaque hemorrhage, larger maximum percentage of lipid-rich necrotic core, and larger maximum wall thickness. Although current limitations of MRI restrict its potential in the coronary vasculature, these data support the contention that features of plaque composition alone may provide value in the assessment of risk of future events.

Another noninvasive technology gaining clinical acceptance is computed tomography (CT) angiography, used primarily for the detection of calcium; however, recently it has been shown that it may be useful in detecting the plaques that may be responsible for ACS. In a recent report, Motoyama examined 2 important CT characteristics of culprit lesions, plaque remodeling index and low attenuated plaques, prospectively in 45 patients for 27±10 months. Of the 45 patients having combined features of plaque remodeling and low attenuated plaque, 10 (22.2%) developed plaque rupture during follow-up versus 1 patient (3.7%) in whom only a single feature was detected. In contrast, of the 820 patients who were negative for both features, only 4 (0.5%) developed ACS. Therefore, patients with coronary lesions exhibiting positive remodeling and low attenuation on CT angiography were considered at higher risk for ACS compared with patients having lesions without these characteristics.
sidering the significant amount of radiation exposure to the patient generated by this modality, before CT screening could be advised, one would need to replicate these findings in a much larger cohort to demonstrate their reproducibility in predicting ACS.

More recently, intracoronary optical coherence tomography (OCT), a technique that measures backscattered light, or optical echoes, derived from an infrared light source directed at the arterial wall, has been proposed as a high-resolution analogue of intravascular ultrasound. The favorable resolution capabilities of 10 μm, validated ex vivo, allow superior definition on the order necessary to resolve thin fibrous caps, macrophages, and necrotic cores. One clinical study reported the detection of TCFAs by OCT with frequencies of 72% for acute MI, 50% for ACS, and 20% for ACS with stable angina. Another interesting study by Tanaka examined 43 consecutive ACS patients who presented with ruptured plaques diagnosed by OCT imaging. On the basis of the level of physical activity at onset of ACS, patients were stratified into a rest and exertion group. The data regarding thickness of the ruptured fibrous cap in the exertion group was significantly higher than that in the rest-onset group (rest onset: 50 μm [interquartile median, 15 μm]; exertion: 90 μm [interquartile median, 65 μm]; P<0.01). These results call into question the paradigm that a standardized morphological set of criteria will be able to identify vulnerable plaque in all patients.

Although OCT is a promising technology, one major limitation is the attenuation of signal intensity by blood necessitating prolonged, proximal occlusions to screen long arterial segments. In light of this, further developments in OCT technology have led to second-generation instruments or optical frequency-domain imaging, a form of OCT capable of acquiring images at much higher frame rates, which enables rapid, 3-dimensional imaging of long coronary segments.

Another option that has recently emerged at the forefront of the cardiovascular imaging field is the identification of putative molecular and cellular targets to illuminate critical processes in the transition from TCFA to rupture. In particular, fluorescence imaging using near-infrared fluorescence has been used to detect proinflammatory and destructive cysteine and serine proteases implicated in plaque rupture, using activated reporter molecules. Putative targets have included components of plaque neoangiogenesis, as well as other elements associated with lesion instability, including inflammation.

In addition, a variety of magnetic nanoparticles or superparamagnetic imaging agents have been developed to detect inflammation in the atherosclerotic plaque. Nanoparticles are engulfed by phagocytic cells and accumulate in resident macrophages of the lesion. One advantage to magnetic nanoparticles is that they are detectable by high-resolution MRI or near-infrared fluorescence. Studies along these lines have focused on imaging vascular cell adhesion molecule-1 in murine models and human plaques. Although proof-of-concept experiments in animal models support the feasibility of such technologies, the major question confronting the field is which biomarkers are the best indicators in the transition from TCFAs to rupture?

Proteolysis and Plaque Imaging

The release of matrix proteases, such as metalloproteinases (MMPs) or cathepsin-associated proteases, may represent another viable target for imaging, considering their role in fibrous cap thinning and plaque destabilization. Typical normal arteries do not possess active MMPs, although they are expressed by macrophages in human atherosclerotic plaques. One limitation to the approach of proteolytic markers for imaging studies is low number of cells (predominantly macrophages) expressing MMPs located in specific regions of the plaque, such as shoulder areas. Therefore, the localization of proteolytic factors would perhaps require sensitive imaging technologies. Despite this potential limitation, several studies have shown increased proteolytic activity associated with macrophage infiltration of plaques using noninvasive techniques, mostly involving radiolabeled molecules targeted at proteases developed for single-photon-emission computed tomography and positron-emission tomography.

Alternative and perhaps more sensitive approaches have also been explored involving the targeting of processes responsible for upregulation of proteolytic activity or near-infrared fluorescent molecular imaging using gelatinase-activated probes to detect the enzymatic activity of MMPs. The first application of this technology was recently reported in carotid plaques examined ex vivo with a MMP-sensitive fluorescent probe, where hot spots corresponding to proteolytic activity of MMP-9 were identified at the origin of the internal carotid artery and at the level of the common carotid bulb.

Catheter-Based Near-Infrared Spectroscopy and Assessment of Lipid Burden

In a recent study from the laboratory of Dr Muller, a novel catheter-based near-infrared spectroscopy system was used to identify lipid burden in human coronary plaques at autopsy with correlative studies by histology. Using an ex vivo model of human coronary segments under simulated pressure, temperature, and flow, lipid core plaque was identified in 115 of 2649 (4.3%) histological sections from 51 validation hearts. The lipid core burden index detected the presence or absence of any fibroatheroma with an area under the curve of 0.86 (95% CI, 0.81 to 0.91) based on receiver-operating characteristics analysis of lipid core index values and histology truth. Moreover, a retrospective analysis of lipid core burden index conducted in extreme artery segments with either no or extensive fibroatheroma yielded an area under the curve of 0.96 (95% CI, 0.92 to 1.00), thus confirming the accuracy of spectroscopy in identifying plaques with diverse lipid content, in an ideal setting. Thus, these preliminary data suggest the potential of an invasive technology to assess lipid core burden, which may be used as an adjunct to
diagnostic angiography or intravascular ultrasound for risk stratification and treatment of patients with ACS.

Involvement of Intraplaque Hemorrhage in the Critical Transition From TCFA to Rupture

Clearly, fibrous cap thickness, necrotic core size, and positive remodeling are critical morphological features that distinguish TCFA from ruptures from earlier progressive lesions, defined as pathological intimal thickening or fibroatheromas. During plaque development, the fibrous cap becomes a functional structure distinct from underlying areas of necrosis, where it harbors the highly thrombogenic contents of the necrotic core. Data from our laboratory provided evidence that repeated intraplaque hemorrhage is a contributing factor to necrotic core expansion, mainly considering that red blood cell membranes serve as a potent source of free cholesterol.54 Based on the earlier work from studies by Katz et al in 197677 and later by Felton in 1977,78 there are significant differences in lipid composition in different lesion types, where free cholesterol is higher in disrupted plaques. In our study, glycoprotein-A, a highly expressed transport protein localized to erythrocyte membranes, is commonly present within the necrotic core of advanced coronary atheroma relative to plaques in earlier phases of development.54 The source of hemorrhage is likely leaky vasa vasorum that infiltrate the plaque from the adventitia in response to a hypoxic environment created by increased lesion burden and inflammatory macrophages. This, together with the death of macrophages in the setting of defective phagocytic clearance of apoptotic cells, is thought to contribute to the development of necrotic core (Figure 4). MRI studies of carotid plaques over an 18-month period showed evidence of intraplaque hemorrhage as a contributing factor to necrotic core volume and lesion bulk.79

What to Look for When Imaging the Vulnerable Plaque

Recent expanded morphometric examination of ruptures and TCFA in our laboratory was performed to further identify relevant parameters of plaque rupture, besides cap thickness. In this series of culprit plaques, 65% of ruptures exhibited >75% cross-sectional luminal area narrowing, whereas 35% showed stenosis of <75%. The latter lesions were further subdivided into those with luminal narrowing of 50% to 75% (28% of lesions) and <50% (7% of lesion) (Figure 5; R. Virmani, unpublished data, 2010). For nonculprit vulnerable plaques, 42% of TCFA exhibited >75% cross-sectional luminal narrowing, and 57% showed stenosis <75%. As for ruptures, the latter lesions were similarly divided into those with luminal narrowing of 50% to 75% (40% of lesions), and 17% showed <50% diameter stenosis. In the clinic, a high percentage of potential vulnerable plaques with >75% stenosis, irrespective of morphology, will likely be treated using an invasive strategy, thereby removing the risk for rupture.80 Therefore, the majority of plaques at high risk for rupture potentially amenable to noninvasive treatment are in the target stenosis range of 50% to 70% (ruptures = 28% versus...
Plaque rupture is a primary underlying cause of luminal thrombosis responsible for provoking ACS. Although plaque erosion and calcified nodules are also accountable for coronary thrombi, unlike ruptures precursors to these lesions have not been identified. The TCFA is considered an unstable high-risk plaque with a propensity toward rupture. These lesions, however, are morphologically different from ruptures, as they generally exhibit smaller necrotic cores and intact thin fibrous caps less infiltrated by macrophages; the degree of calcification is also less. The current paradigm designating TCFAs as a prelude to ruptures is primarily supported by autopsy findings, where definitive proof does not exist because of a lack of prospective animal or human data confirming a cause-and-effect relationship. Potential morphological and biological processes that may be helpful for the identification of TCFAs recognized by today’s imaging modalities include necrotic core size, intraplaque hemorrhage, and positive remodeling. Advancing the field, however, will require establishing relevant translational animal models that produce vulnerable plaques at risk for rupture and furthering the development of novel imaging and therapeutic modalities.

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None.

References


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