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Review

Plaque-associated fibroblasts: Key regulators of atherosclerosis pathogenesis and plaque stability

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Abstract. *Background and objective:* Plaque-associated fibroblasts (PAFs) play a crucial role in shaping the plaque's trajectory, either towards stability or instability. The pathological transformation of fibroblasts into myofibroblasts, characterized by increased contractility and secretion, contributes to excessive extracellular matrix (ECM) deposition. The bidirectional crosstalk between fibroblasts and inflammatory cells within the plaque is a crucial aspect. Activated fibroblasts release proinflammatory factors like interleukin-1 (IL-1), activating resident immune cells and facilitating their migration through the plaque microenvironment (PME). Conversely, immune cells produce cytokines such as IL-6, TNF-alpha, TGF-beta, and IL-1beta, stimulating fibroblasts to produce matrix metalloproteinase 1 (MMP1) and collagen deposition. The dynamic interplay among these cells, influenced by genetic predispositions, systemic conditions (hypertension, diabetes), inflammatory states (including COVID-19), and environmental factors (diet, lifestyle), determines the plaque's fate. This review discusses the natural progression of carotid plaque and the evolving concepts surrounding the multiple events underlying vulnerable atherosclerotic lesions. *Method:* Google Scholar, Scopus, and PubMed were searched for manuscripts on PAFs and those reporting the association between PAFs and atherosclerosis. *Conclusion:* Advances in our interpretation of histological images of atherosclerotic lesions may pave the way for novel therapeutic strategies aimed at inhibiting detrimental PAF activity, thereby facilitating further plaque stabilization and preventing severe clinical complications arising from carotid atherosclerotic plaque rupture. (www.actabiomedica.it)

Key words: atherosclerotic plaque, plaque stability, fibroblast activation, vascular inflammation, extracellular matrix remodeling, atherosclerosis pathogenesis, plaque progression, fibroblasts in cardiovascular disease, vulnerable plaque dynamics.

Introduction

Recent evidence has spurred a considerable evolution of theories and concepts regarding the insurgence and progression of atherosclerotic lesions, calling into question multiple previous notions on this complex and partly unknown disease (1). In this changing landscape of atherosclerosis, noninvasive carotid imaging modalities, and novel standardized reporting systems have demonstrated their ability to characterize features of plaque instability as predictors of future adverse events (2). Probing the pathogenesis and histology of atherosclerosis has highlighted the role of the plaque-associated fibroblasts (PAFs), which represent

the major cell population involved in remodeling of the extracellular matrix (ECM) in the atherosclerotic plaque (3). Characterizations of this distinct fibroblast population within the plaque have advanced in recent years, leading to improved identification of multiple PAF subtypes. These subtypes exhibit varied and occasionally contrasting functions that influence the evolution of carotid plaques toward either a stable or unstable state, with severe detrimental clinical consequences including rupture (4). This literature review addresses the evolving concepts of the origin of plaque-associated fibroblast-like cells, focusing on their identification, their crosstalk with the other cell types that characterize the plaque microenvironment (PME), their ability in orchestrating the evolution of the plaque towards a stable or an unstable state.

Natural history of carotid atherosclerotic plaque

The traditional view of the atherosclerotic plaque, the hallmark lesion of atherosclerosis, was primarily centered on lipid accumulation within the intima of arterial vessels. According to this perspective, the cytoplasmic storage of lipids in macrophages was followed by the emergence of extracellular lipids, leading to the formation of confluent masses of lipids and necrotic cells, ultimately resulting in the formation of an atheroma. The migration of smooth muscle cells from the media and their activation into fibroblasts contributed to the development of fibroatheroma, which could subsequently lead to complications such as erosion, ulceration, and thrombosis, culminating in severe clinical consequences (5). In this 'lipid-centric' paradigm of atherosclerosis, high levels of cholesterol, apolipoprotein B, and LDL were considered to play a predominant role in the initiation and progression of atherosclerotic lesions (6). Recent studies utilizing machine learning to understand atherosclerotic lesions have underscored the multifactorial nature of plaque formation. Factors such as endothelial dysfunction, inflammation, proliferation and differentiation of vascular smooth muscle cells, biomechanical and hemodynamic forces, and intra-plaque hemorrhage have been recognized as contributing to plaque development (7,8). Studies aimed at identifying molecular events characterizing the transition from a stable plaque to a vulnerable lesion have identified the reduced number of intra-plaque vascular smooth muscle cells, associated with weakening of the fibrous cap, as a key histological feature of unstable atherosclerotic plaques (9,10). These collective findings prompted Isabel Goncalves and colleagues to propose the plaque vulnerability index for histological analysis of carotid plaques following endarterectomy (11). This study underscores the importance of accurate histological analysis of plaques removed from carotid arteries by pathologists. Given that atherosclerosis is a systemic disease, individuals with a high vulnerability index in carotid plaques should be considered at higher risk of developing unstable plaques in other vascular territories, including the coronary tree. Consequently, these individuals should undergo stratification and stringent follow-up to prevent severe clinical events.

Fibroblasts in health and disease

Fibroblasts, the architecturally fundamental cells of connective tissues, play a key role in maintaining tissue homeostasis. These versatile cells, ubiquitous throughout the human body, are paramount in the synthesis of ECM components, thus providing structural and biochemical support to surrounding cells. Fibroblasts are cells of mesenchymal origin and represent one of the most difficult mesenchymal cell types to define, due to their heterogeneity, with multiple subtypes typical of different tissues and organs, Moreover, fibroblasts are characterized by marked plasticity, by the ability to transform into myofibroblasts when activated, and by their dynamic behavior, with the ability to acquire different functions according with the environment in which they are embedded (4). Fibroblasts from different tissues and anatomical sites show different phenotypes, and different functions and are characterized by distinct transcriptional patterns regarding genes involved in lipid metabolism, ECM synthesis and remodeling, and in molecular pathways involved in cell proliferation, migration, and fate determination (12). Multiple HOX genes, encoding multiple transcription factors, are differently expressed in fibroblasts localized in different anatomical sites. In adulthood, fibroblasts maintain the HOX gene expression patterns established during embryogenesis, underlying the

positional memory of fibroblasts through the whole life (12). In a healthy state, fibroblasts are instrumental in wound healing, demonstrating remarkable plasticity (13). Their ability to transition between quiescent and active states is essential for the reparative processes following tissue injury. Activated fibroblasts may promote the activation of macrophages, significantly improving wound healing (14). Moreover, during wound healing, fibroblasts may recruit and activate innate immune cells, showing a peculiar versatility in the healing machinery, that induced some authors to define them as "the choreographers of wound healing" (15). However, the role of fibroblasts extends beyond tissue repair and maintenance. In the context of pathological conditions such as atherosclerosis, fibroblasts exhibit a transformative potential that can be both beneficial and detrimental. The pathological transformation of these cells into myofibroblasts, a contractile and secretory variant of fibroblasts, can lead to excessive ECM deposition, culminating in fibrosis (16). This aberrant fibroblastic activity is a hallmark of various chronic diseases, including atherosclerosis, contributing significantly to morbidity and mortality (17). Table 1 summarizes the role of fibroblast in healthy patients and during atherosclerosis.

In healthy arterial vessels, fibroblasts may be found in the adventitia, which represents the most complex layer of blood vessels, being characterized by a heterogeneous composition of cells including fibroblasts, resident immune cells such as dendritic cells, macrophages, pericytes, stem/progenitor cells, adrenergic nerve cells and small blood vessels with endothelial cells, better known a vasa vasorum (18). Adventitial fibroblasts show marked differences regarding morphology, (spindle, or epithelioid), size, function and activity. They are the first cells of the arterial vessel to become activated in response to multiple triggers, including inflammatory and hormonal stimuli, environmental stress, hypoxia, ischemia, and hypertension. In physiology, a major function of fibroblasts is the synthesis of ECM components, including collagens, proteoglycans, elastin, fibronectin, microfibrillar proteins, and laminins. They also secrete lysil oxidase, multiple MMPs, and MMP inhibitors, actively remodeling ECM structure and architecture during development (19). Activated fibroblasts increase the production of multiple ECM proteins, including collagen, elastin, and proteoglycans, Moreover, when activated, adventitial fibroblasts may regulate the growth of vasa vasorum through the production of vascular endothelial growth factor (VEGF), transforming growth factor beta (TGF-beta), platelet-derived growth factor (PDGF) and monocyte chemoattractant protein 1 (MCP1). Wnt signaling pathway also plays a key role in regulating fibroblasts' proliferation, migration, and their ability in ECM deposition during development (20). During embryonic development, fibroblasts play crucial roles in multiple fields, including ECM homeostasis, secretome (production of cytokines, adipokines, growth factors), mechanical forces (contraction, directional migration, ECM polarization), metabolism (glycolysis, lipid metabolism), regulation of innate immunity (macrophage polarization, antigen presentation through differentiation toward an antigen-presenting cell phenotype), positional/niche information (signaling center for stem cells), tissue synthesis (stroma formation in organ morphogenesis), progenitor function (progenitor self-renewal of fibroblasts, mesenchymal lineage maintenance) (20) (Figure 1).

The plaque microenvironment (PME)

The histological composition of the atherosclerotic plaque typically consists of the following elements: a necrotic lipid core, extracellular lipids, cholesterol deposits, and multiple cell types including smooth muscle cells, macrophages, monocytes, lymphocytes, fibroblasts, all capped by a layer of fibrous tissue composed by a dense connective ECM. Further elements possibly present in the PME are macro- and microcalcifications, intraplaque hemorrhages, rupture of the fibrous cap, ulceration, and thrombosis (21–23). Occasionally, plasma cells may characterize the PME (23). The PME in atherosclerotic lesions presents as a complex and dynamic cellular theatre, where diverse cellular constituents engage in a continuous and intricate interplay. This microenvironment is not merely a passive collection of cells and extracellular matrix; rather, it is a highly active and interactive locale, crucial for the progression or stabilization of the atherosclerotic

Table 1. The Role of Fibroblasts in Healthy States and Atherosclerosis

Aspect	Details	Functional Impact
Basic Role	Architecturally fundamental cells of connective tissues, vital for maintaining tissue homeostasis. Involved in ECM synthesis, remodeling, and managing volume and pressure of interstitial fluid.	Provide structural and biochemical support to cells and manage ECM dynamics.
Origin and Diversity	Cells of mesenchymal origin, heterogeneous with multiple subtypes in different tissues and organs. Characterized by plasticity and dynamic behavior, capable of transforming into myofibroblasts and adapting to environmental conditions.	Different functions and behaviors based on tissue location and environmental conditions.
Genetic Expression	Distinct transcriptional patterns in lipid metabolism, ECM synthesis, and cell proliferation. Multiple HOX genes encoding transcription factors are expressed differently in fibroblasts from different anatomical sites.	Determine the distinct phenotypes and functions of fibroblasts in various tissues.
Role in Wound Healing	Demonstrates remarkable plasticity in wound healing. Can transition between quiescent and active states, promoting macrophage activation and recruiting innate immune cells.	Essential for reparative processes and effective wound healing.
Pathological Conditions	Transformative potential in conditions like atherosclerosis, leading to excessive ECM deposition and fibrosis.	Can be both beneficial and detrimental, contributing to chronic diseases.
Role in Arterial Vessels	Located in the adventitia of healthy arterial vessels, showing marked differences in morphology, size, function, and activity. First to activate in response to stimuli like inflammation and stress.	Regulate arterial vessel response to various stimuli.
ECM Synthesis & Remodeling	Synthesize ECM components (collagens, proteoglycans, etc.) and secrete enzymes for ECM remodeling. Activated fibroblasts increase production of ECM proteins and regulate the growth of vasa vasorum.	Actively remodel ECM structure and contribute to development.
Embryonic Development Roles	Involved in ECM homeostasis, secretome production, mechanical forces, metabolism, regulation of innate immunity, positional/niche information, and tissue synthesis. Act as a signaling center for stem cells and maintain mesenchymal lineage.	Play key roles in organ morphogenesis and various embryonic developmental processes.

plaque (Table 2). Atherosclerosis is a chronic disease driven by inflammation of the arterial vessels induced by multiple factors including low-density lipoproteins, reactive oxygen species, infections, mechanical stresses, and chemical insults. Each of the cell types in the ECM contributes uniquely to the plaque's evolution. Endothelial cells, lining the inner walls of arteries, are initial responders to atherogenic stimuli, while smooth muscle cells, migrating from the media to the intima, contribute to plaque formation and stability. Immune cells, such as monocytes/macrophages, T lymphocytes, B lymphocytes, plasma cells, and dendritic cells, infiltrate the plaque and partake in inflammatory responses and fibrosis, influencing plaque progression and potential rupture. Active resident fibroblasts, myofibroblasts, and smooth muscle cells are deputed to a continuous remodeling of the extracellular matrix of the plaque, under the control of paracrine and autocrine signaling from the immune cells infiltrating the PME. The crosstalk between fibroblasts and

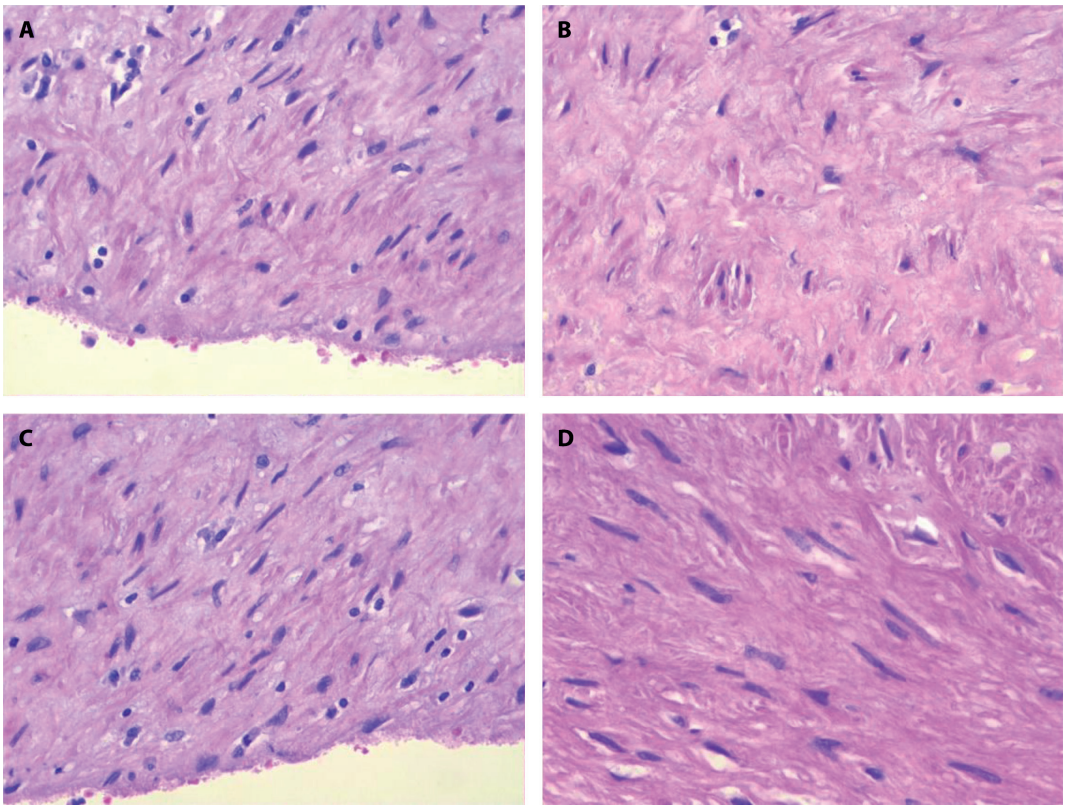

Figure 1. Histological appearance of plaque-associated fibroblasts. a) spindle-shaped fibroblasts embedded in a fibro-mycoid matrix. b) sea-shaped or triangular activated myofibroblasts with a hypereosinophilic cytoplasm scattered in a fibrous matrix. c) spindle fibroblasts with elongated nuclei intermingled with lymphocytes and monocytes. d) at high power, spindle-shaped fibroblasts show vesicular chromatin, nuclei with smooth extremities, and peri nuclear vacuoles, features suggestive of a possible origin from smooth muscle cells migrating from the tunica media. All figures in the panel are the intellectual property of the authors.

inflammatory cells inside the plaque is bidirectional: activated fibroblasts produce multiple proinflammatory factors, including interleukin-1 (IL-1), able to activate resident immune cells, favoring their migration through the PME. On the other hand, immune cells produce multiple cytokines, such as IL-6, TNF-alpha, TGF-beta, and IL-1beta, which activate the fibroblasts of the PME stimulating their production of matrix metalloproteinase 1 (MMP1) and collagen deposition (3). Within this milieu, PAFs play a critical role. They are not mere bystanders but active participants who significantly influence the PME's characteristics. Through their interactions with other cell types and their ability to remodel the extracellular matrix, PAFs can either promote a stabilizing fibrotic response or contribute to plaque vulnerability. Gender- and age-related differences have been reported to influence the composition of the PME. Women tend to develop plaques richer in ECM components, with a smaller lipid-rich necrotic core as compared to men. On the contrary, plaques in men show a higher prevalence of lipid-rich necrotic cores, intraplaque hemorrhage, lower collagen deposition, and more frequent microcalcifications of the PME, all markers associated with higher plaque vulnerability (24). Older age has been associated with rupture-prone plaques, with a PME characterized by lower collagen deposition, lower content of smooth muscle cells, and higher presence of microcalcifications. Interestingly, the PME of older women changes when compared to that typical of young women: in postmenopausal age, plaques show more rupture-prone characteristics of their PME,

Table 2. Summary of the Plaque microenvironment

Component/Aspect	Details	Impact on Atherosclerosis
Histological Composition	Consists of a necrotic lipid core, extracellular lipids, cholesterol deposits, various cell types (smooth muscle cells, macrophages, monocytes, lymphocytes, fibroblasts), all capped by a fibrous tissue layer composed of dense connective ECM. May also contain macro-/micro-calcifications, intraplaque hemorrhages, fibrous cap ruptures, ulceration, and thrombosis.	Defines the structural and functional characteristics of the plaque, influencing its stability or vulnerability.
Dynamic Cellular Theatre	A complex interplay among diverse cellular constituents, including endothelial cells, smooth muscle cells, immune cells, monocytes/macrophages, and fibroblasts.	Cellular interactions crucial for plaque progression or stabilization.
Inflammation and Fibrosis	Inflammation drives the disease, with fibrosis compensating for arterial wall injury. Immune cells and fibroblasts produce proinflammatory factors and matrix metalloproteinases (MMPs), contributing to ECM remodeling.	Chronic inflammation and fibrotic response significantly influence plaque progression and potential rupture.
Role of Macrophages	Produce MMPs and tissue inhibitors of MMPs (TIMPs), as well as TGF-beta. Balance between MMPs and TIMPs is crucial for ECM deposition and degradation, affecting myofibroblast differentiation.	Critical in ECM remodeling, influencing plaque stability.
Endothelial Cell Function	Undergo endothelial-to-mesenchymal transition (EndMT), acquiring a migratory phenotype and differentiating into myofibroblastic-like cells producing collagen and ECM components.	Contribute to collagen deposition and ECM remodeling, affecting plaque characteristics.
Growth Factors	Produced by smooth muscle cells, monocytes, macrophages, and endothelial cells, including PDGF, TGF alpha and beta, and FGF. These factors are mitogenic for smooth muscle and endothelial cells, promote angiogenesis, and facilitate ECM deposition.	Mediate crucial processes in plaque development and stability.
Gender- and Age-Related Differences	Women tend to develop plaques richer in ECM components, while men show plaques with higher prevalence of lipid-rich necrotic cores and other vulnerability markers. Older age associated with rupture-prone plaques. Postmenopausal women show more rupture-prone plaque characteristics compared to younger women and men.	Gender and age significantly influence plaque composition and vulnerability.

compared to younger women and young men (25). All these data taken together indicate a relevant effect of menopause on plaque morphology and, in particular, on the remodeling of the PME of the atherosclerotic lesions, suggesting a strong interaction between aging and gender on the evolution of the plaque (26). The major role of aging and gender in PME composition of the atherosclerotic plaques has been recently confirmed by Benavente (27) and coworkers in a study focused on the expression of female gene networks in myofibroblastic-like smooth muscle cells in vulnerable plaques (27). In this study, young women presenting with coronary artery disease (CAD) often showed plaques characterized by a fibrous PME and were protected against severe cardiovascular consequences. At older ages, in their 70s', the PME changed significantly, and the incidence of CAD in women surpassed that of men of the same age.

The roles of fibroblasts in atherosclerosis

In atherosclerosis, a disease characterized by the thickening and hardening of arterial walls, fibroblasts assume a specialized role. The plaque-associated fibroblasts (PAFs), situated within atherosclerotic lesions, are pivotal in plaque formation and progression. These cells, through their interplay with the ECM, influence

the stability of the atherosclerotic plaque. Their activity can dictate the transition of the plaque towards a stable or unstable state, thereby impacting the likelihood of plaque rupture and subsequent cardiovascular events. Another player in the formation of the neointima is represented by the fibroblasts physiologically localized in the adventitia, at the periphery of the arterial wall, and away from the lumen. These cells, seen for a long time as innocent bystanders, in recent years have been reinterpreted as co-authors of the vascular remodeling typical of the atherosclerotic process and active participants of the neointima formation(28). Adventitial fibroblasts, when activated, transform into myofibroblasts, which may migrate into the neointima where they upregulate the production of multiple proinflammatory cytokines and chemokines, of growth factors, proteolytic enzymes, and ECM proteins, remodeling extensively the PME and recruiting immune cells inside it (4).

The origins of PAFs

Myofibroblasts of the plaque may have multiple and different origins. PAFs may derive from a) smooth muscle cells migrating into the neointima from the underlying tunica media; b) adventitial fibroblasts, migrating from the adventitia through the tunica media and into the neointima (28,4); c) endothelial cells through the process of endothelial-to-mesenchymal transition (29,30); d) quiescent residential cells of the plaque, including fibrocytes and fibroblasts (31); e) bone marrow-derived circulating fibrocytes (32,33); f) monocytes/macrophages (34); g) peri-adventitial adipocytes may reenter cell cycle and produce new ECM-making fibroblasts (20); h) perivascular adipose tissue (PVAT) stem/progenitor cells, normally localized around the adventitia (35,3). In recent years, the source of mesenchymal cells in the atherosclerotic plaque has been under scrutiny. Many studies were focused on the endothelium, as an important source of PAFs. In the complex process leading to the insurgence of the atherosclerotic plaque, endothelial cells can acquire a mesenchymal phenotype through the process called endothelial-to-mesenchymal transition (EndMT). This process originates “hybrid cells”, mesenchymal cells of endothelial origin, which detach from the adjacent endothelial cells and migrate into the PME (36). Endothelial cells undergoing mesenchymal transition progressively lose their characteristics and gain a mesenchymal phenotype, with a rearrangement of actin filaments that facilitates the acquisition of a motile phenotype and primes their migration into the plaque (37). Once arrive at the plaque, these hybrid cells may differentiate into myofibroblasts, contributing to the fibroproliferative expansion of the plaque (30). The shear stress occurring in some peculiar tracts of the arterial bed has been indicated as the trigger of endMT in a peculiar tract of the arteries, through the transcription factor Snail (38). Moreover, the extent of endMT has been associated with plaque instability, giving this process a negative prognostic significance (39). According to this hypothesis, further studies suggested the interpretation of endMt as a type of endothelial dysfunction, probably induced by the shear stress, that may cause vascular remodeling, a major determinant in the insurgence of the atherosclerotic plaque (40). More recent studies indicate endMT as a physiological process that plays a major role during development, a complex set of interactions of multiple molecular mechanisms active before birth, silenced in childhood, and which is reactivated in some chronic diseases, including atherosclerosis (41). Table 3 summarizes the role of fibroblasts and PAFs in atherosclerosis.

PAF subtypes

The plasticity of fibroblasts associated with the atherosclerotic plaques and their multiple origins from different cell types are the main factors at the basis of the existence of multiple subtypes of PAFs, with different roles in plaque insurgence and evolution. Regarding adventitial fibroblasts, at least four subsets have been identified yet. Among them, Sca1-positive fibroblasts have been associated with the ability to migrate towards the neointima, whereas Gli1-positive cells show the ability to differentiate into osteoblasts, significantly contributing to the calcification of the plaque (31). Regarding myofibroblasts, the activated form of fibroblasts, according with their immunohistochemical phenotype, five subtypes have been identified utilizing a panel of four antibodies: 1) V-type myofibroblasts, characterized by the expression of vimentin;

Table 3. The Role of Fibroblasts and Plaque-Associated Fibroblasts in Atherosclerosis

Aspect	Description	Origins/Types	Functions
Role in Plaque Formation	PAFs influence plaque stability, dictating transition towards stable or unstable states.	Myofibroblasts from smooth muscle cells, adventitial fibroblasts, endothelial cells, quiescent residential cells, bone marrow-derived fibrocytes, monocytes/macrophages, adipocytes, PVAT stem/progenitor cells.	Dictate plaque stability and likelihood of rupture.
Endothelial-to-Mesenchymal Transition (EndMT)	EndMT produces hybrid cells that may differentiate into myofibroblasts in the plaque. Triggered by factors like shear stress.	Endothelial cells undergoing mesenchymal transition.	Contribute to fibroproliferative expansion of the plaque. Linked with plaque instability.
PAF Subtypes	Multiple subtypes of PAFs exist due to fibroblasts' plasticity and multiple origins.	At least four subsets of adventitial fibroblasts, including Sca1-positive and Gli1-positive cells.	Different roles in plaque onset and evolution.
Immunohistochemical Markers	Identification of fibroblasts in the PME using markers like vimentin, PDGFR-alpha, FAP, FSP1, DCN, COL1A1, fibronectin, prolyl-4-hydroxylase, discoidin-domain receptor. Differentiation from smooth muscle cells.	Markers include vimentin, PDGFR-alpha, FAP, FSP1, DCN, COL1A1, fibronectin, prolyl-4-hydroxylase, discoidin-domain receptor. Smooth muscle cells show markers like ACTA2, MYOCD, MYH11, PDGF-beta, KLF4, MMP2, CNN1.	Identification and differentiation of fibroblasts in the PME.
Functions of PAFs	PAFs uptake oxLDL, involved in calcification, and responsible for fibrosis and remodeling. They regulate inflammation through cytokine production and maintain the structural integrity of the PME.		Uptake oxLDL, contribute to calcification, responsible for fibrosis, remodeling, and inflammation regulation.

2) VD-type, expressing vimentin and desmin; 3) VAD-type, expressing vimentin, desmin and alpha-smooth muscle actin; 4) VA-type, immunoreactive for vimentin and alpha-SMA; 5) VM-type, characterized by the expression of vimentin and myosin (17).

Functions of PAFs in the definition of the architecture of the plaque

Plaque myofibroblasts may uptake oxLDL, a key event for the insurgence of the lipid-rich necrotic core in the PME of the plaque. PAFs are also involved in the calcification of the plaque, leading to vascular calcification, micro- and/or macro-calcifications of the ECM of the PME, favoring the occlusion of the lumen or, alternatively, leading to the rupture of the plaque, followed by thrombosis. PAFs are also the main one responsible for fibrosis and remodeling, the processes of compensation for tissue injury caused by chronic inflammation occurring in the PME. In the presence of ongoing unresolved inflammation, persisting signaling for myofibroblastic activation causes an excess in ECM deposition in the PME, followed by disorganization and thickening of the plaque. The proper balance between MMPs and TIMPs is crucial for the normal deposition and degradation of the ECM of the PME (3). Another key function of fibroblasts inside the plaque is the regulation of inflammation in the PME, through the production of excess cytokines and chemokines. The maintenance of the structural integrity of the PME is also a key function of PAFs, through the production of proteolytic enzymes and the modulation of collagen deposition in the PME.

Future directions

New advances in our interpretation of the histological images of atherosclerotic lesions might open avenues to new therapeutic strategies aimed at inhibiting detrimental smooth muscle cell transition towards an aggressive myofibroblastic phenotype, allowing a further stabilization of the plaque and preventing the severe clinical complications caused by the rupture of carotid atherosclerotic plaques (43-45). Understanding the dual nature of fibroblasts in health and disease opens up new therapeutic avenues. Targeting fibroblast activity, particularly in the context of atherosclerosis, presents a promising strategy for mitigating the progression of the disease. Modulating PAF activity could potentially stabilize atherosclerotic plaques, reducing the risk of rupture and the severe clinical complications that follow. Therefore, further research into the specific roles and mechanisms of fibroblasts, particularly PAFs in atherosclerosis, is crucial for the development of effective therapeutic interventions. Clarifying the multiple subtypes of PAFs and their multiple functions, including the collagen deposition and remodeling of the ECM in the different phases of the atherosclerotic plaque evolution holds remarkable therapeutic potential. Given the major role played by PAFs in monitoring the ECM composition and architecture, it is conceivable that the induction of fibroblasts acting inside the plaque to re-establish a correct MMP/TIMP balance, through the use of MMP inhibitors, might stabilize the plaques, avoiding a progression towards clinical detrimental consequences.

Conclusion

The evolution of our understanding and interpretation of histological images portraying atherosclerotic lesions presents a compelling opportunity for the development of pioneering therapeutic modalities. By specifically targeting the activity of PAFs, these emerging strategies aim to fortify plaque stability and mitigate the devastating consequences associated with the rupture of carotid atherosclerotic plaques. Such interventions hold substantial promise in revolutionizing patient care by addressing critical cellular mechanisms underlying plaque progression and vulnerability.

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