REVIEW ARTICLE

Acquired dysfibrinogenemia in atherosclerotic vascular disease

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KEY WORDS

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ABSTRACT

Acquired qualitative abnormalities of fibrinogen molecules, termed acquired dysfibrinogenemia, have been demonstrated in several disease states mostly related to prothrombotic tendency, including multiple myeloma and liver disease. Fibrin is abundant in atherosclerotic plaques. Altered plasma fibrin properties, reflected usually by reduced clot permeability and impaired fibrinolysis, have been reported in patients with acute or prior myocardial infarction, ischemic stroke, and peripheral artery disease. Moreover, prothrombotic clot phenotype has been observed in patients with previous no-reflow phenomenon and stent thrombosis. Growing evidence indicates that acquired dysfibrinogenemia contributes to the progression of atherosclerotic vascular disease and the occurrence of its thrombotic manifestations. The review summarizes current knowledge on the links between fibrin clot phenotype and atherosclerotic vascular disease and describes a wide spectrum of cardiovascular risk factors as modifiers of fibrin network characteristics.

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Fibrin clot formation and degradation Fibrin is generated in a process mediated by thrombin from fibrinogen, a plasma 340-kDa dimeric glycoprotein, which is a hexamer composed of 3 paired polypeptide chains $(A\alpha, B\beta, \gamma)_2$ linked by 29 disulfide bonds. Fibrinogen contains 3 main structural regions connected by α-helical coils: a central E domain with N-termini of all 6 polypeptide chains (with fibrinopeptides A, B) and 2 outer D domains with C-termini of Bβ and γ chains.[1] Fibrin clot formation, being the final step in blood coagulation, results from a series of rapid events initiated by thrombin cleavage of the Aα- and Bß-chains of fibrinogen. Thrombin-mediated release of FPA and, much slower, FPB from the amino-termini of the Aα- and Bβ-chains of fibrinogen, respectively, results in the formation of Fn monomer with the structure $(\alpha, \beta, \gamma)_2$. When protofibrils grow sufficiently long, they undergo lateral aggregation to make a fibrin fiber.[1,2] Impaired lateral aggregation usually yields clots made up of thin fibers with many branch points.[3] Fibrin clot resistance to enzymatic degradation is largely determined by covalent cross-linking catalyzed by activated FXIII of transglutaminase activity.[4] The proper balance between fibrin formation, its cross-linking, and degradation is necessary to protect the vascular system from excess blood loss and also from obstructed blood flow.

Dissolution of a fibrin clot results is mediated by the interaction of tissue plasminogen activator (tPA) and plasminogen on a fibrin surface. The presence of fibrin greatly accelerates by at least 2 orders of magnitude plasmin generation catalyzed by tPA, which is limited proteolysis of plasminogen.[5] Plasmin bound to fibrin is protected from the action of α_2-antiplasmin. The thrombin-activatable fibrinolysis inhibitor down-regulates plasminogen activation and fibrinolysis.[5] Fibrin structure directly affects the rate of fibrinolysis.[6] In vivo fibrinolysis appears to be a heterogenous process, locally extremely rapid while other sites remain un-lyzed, but determinants of this process are not fully elucidated. Plasmin-mediated fibrin degradation leads to the release of specific cross-linked fibrin degradation products, termed D-dimers, which are a sensitive marker of in vivo fibrin formation and lysis.

Dysfibrinogenemia Dysfibrinogenemias are infrequent congenital or acquired qualitative abnormalities of fibrinogen that are most commonly

diagnosed in adults. Congenital dysfibrinogenemia is caused by heterozygosity for a mutation within any of the 3 fibrinogen chain genes (4q28.1, 4q28.2, and 4q28.3 for *FGG*, *FGA*, and *FGB*, respectively), most commonly in the first 2 genes. Congenital dysfibrinogenemia is associated predominantly with defective fibrinopeptide release and/or with retarded fibrin polymerization, and is in most cases detected incidentally.[7] Clinically overt manifestations of dysfibrinogenemia involve bleeding, usually related to trauma, surgery, or childbirth (in 20% of cases) and/or thrombosis (in 25% of cases). Less common manifestations of congenital dysfibrinogenemia involve an increased risk of miscarriages, umbilical cord bleeds, and prolonged wound healing.[7] In a vast majority of congenital dysfibrinogenemia, there is a discrepancy between the levels of clottable fibrinogen (determined most frequently using the Clauss method) and immunologically measured fibrinogen. Functional fibrinogen levels are typically lower than its antigen concentration, with fibrinogen activity-antigen ratio being in most cases approximately 1:2. Thrombin time is commonly prolonged in congenital dysfibrinogenemia.[7]

Acquired dysfibrinogenemia is a rare abnormality that can be observed in a subset of patients diagnosed with liver disease (hepatoma, chronic active hepatitis, cirrhosis, and isolated obstructive jaundice), multiple myeloma, autoimmune disorders, and in some cases of cancer; dysfibrinogenemia may also be induced by medications (e.g., isotretinoin, glucocorticoid, antileukemic agents).[7] This type of dysfibrinogenemia is typically associated with normal fibrinogen activity and antigen levels as well as results of routine coagulation tests, including thrombin time. Acquired dysfibrinogenemia is usually associated with a prothrombotic state and largely results from interactions of fibrinogen molecules with other plasma proteins, e.g., paraproteins, and posttranslational modifications of fibrinogen molecules, e.g., oxidation, which have not been well described yet.[7] Assessment of such abnormalities, which are dynamic by nature and might be transient, require particular methodology to show subtle alterations in fibrin clot structure and function.

Measures of clot properties Fibrin clot structure and function can be assessed using several measures, including: 1) clot permeability, or Darcy constant, termed K_s (an indicator of the pore size), calculated based on the volume of a buffer flowing through a fibrin gel in a given time period; 2) the lag phase by turbidimetry that reflects the time to the start of lateral fibril aggregation; 3) maximum absorbancy of the growing clot that reflects an average fibrin fiber size and the number of protofibrils per fiber.[8] Clot turbidity is related to the number of fibrin fibers, their thickness, and branching points, as well as to the uniformity of fiber distribution.[9]

Imaging techniques used to assess fibrin clot structure include scanning or transmission electron microscopy and also confocal microscopy.[10] Scanning electron microscopy represents the most commonly used technique. It allows the measurement of fiber diameter, pore size, and branching angles. However, this technique requires fixing a clot mostly by permeating it with glutaraldehyde solution with subsequent dehydration.

Fibrin clot properties can be studied in solutions of purified or recombinant fibrinogens and also in citrated plasma upon addition of varying concentrations of exogenous thrombin and calcium. From a pathophysiological point of view, plasma-based assays appear to be closer to the in vivo situation where fibrin(ogen) is subject to modulatory effects of other circulating proteins or other molecules. It is known that fibrin network formed from citrated plasma is composed of thicker fibers compared with that formed from purified fibrinogen.[11]

Viscoelastic properties of clots, such as the dynamic storage modulus, the loss modulus, the tau delta (an indicator of the clot irreversible deformation), and clot stiffness, can be assessed using a Plazek torsion pendulum. Thromboelastography evaluates a global function of hemostasis and also describes elastic properties of the clot during fibrin polymerization.[12]

There is a correlation between fibrin structure and efficiency of fibrinolysis. Clots that are formed from thin, tightly packed fiber strands with small porosity and increased branching are more resistant to lysis.[13] In most clot lysis assays, recombinant tPA at varying concentrations, ranging usually from those encountered during lytic therapy in vivo to 10-fold higher values, is added simultaneously with thrombin. Another approach without addition of exogenous thrombin uses coagulant reactions triggered by tissue factor (TF) in the presence of phospholipid vesicles and plasma, which results in much longer lysis time (usually from 50 to 100 minutes).[14] Resistance to plasmin-mediated proteolysis can be assessed by measurement of the time required for a decrease of the absorbancy of a fibrin clot by 50% of the peak value. Using plasma fibrin clots, prepared as in the permeation assays, and a buffer containing recombinant tPA, determination of D-dimers in the buffer percolating through the fibrin gel estimates the kinetics of fibrin degradation prior to collapse of a clot.[15] Dynamic confocal microscopy has confirmed slower fibrinolysis in tight fiber meshworks compared with clots of thicker and looser fibers, despite the fact that individual thicker fibers are lysed at a slower rate than the thin ones are.[6,13] This observation could be explained by the fact that fewer thicker fibers can be more rapidly digested at the local sites also due to increased tPA binding, and the progressive fiber aggregation may locally enhance lysis because cleavage proceeds laterally.[6,16] Results of different clot lysis assays are weakly correlat-

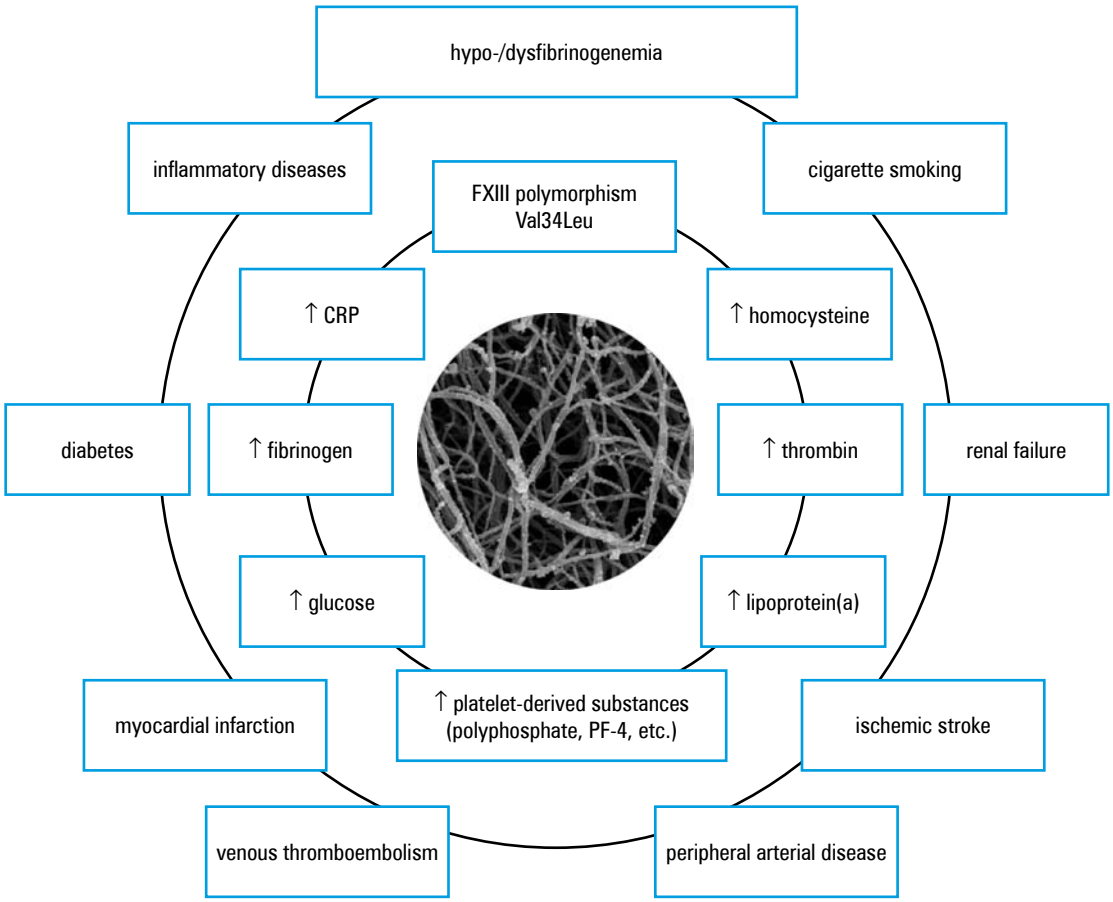

FIGURE Disease states linked with the presence of prothrombotic fibrin clot phenotype, particularly reduced clot permeability and susceptibility to lysis. Apart from atherosclerotic vascular diseases, other conditions associated with increased risk of thrombotic events have been shown to highlight similarities in fibrin parameters between various disorders.
Abbreviations: CRP – C-reactive protein, PF-4 – platelet protein 4

ed suggesting that under different experimental conditions the efficiency of lysis differs.

Fibrinogen levels are recognized as the most important modulator of fibrin clot structure, though variation in fibrinogen concentrations explains 18% of the variation in clot permeability.[17] It has been shown that the structure of the clot depends not only on a fibrinogen concentration, but also on the conditions during fibrinogen conversion to fibrin.[6,16] Positively charged molecules, such as calcium, increase fiber diameter, while thrombospondin and elevated immunoglobulin levels exert the opposite effect on fibers.[18,19] In most purified systems, as fiber size increases, the pore size increases. Fibrin networks composed of the thin fiber strands usually have small pores and are more rigid and less permeable. On the contrary, clots formed by thick fiber strands have large liquid spaces, which implies higher permeability and accelerated fibrinolysis, likely due to a more efficient transport of fibrinolytic agents through a fibrin clot.[6,16,18,19]

Fibrin clot properties and cardiovascular risk factors Growing evidence indicates that altered fibrin clot formation and/or degradation occur in atherothrombotic vascular disease and they might be implicated in its development and complications (FIGURE). The alterations of clot characteristics in this pathology, as in other acquired abnormalities, are determined by environmental and to much lesser extent by genetic factors, e.g., the common factor XIII Val34Leu polymorphism.[19,20] One might expect that traditional risk factors contribute to alterations in fibrin clot phenotype. However, data on such associations, except diabetes, are rather scarce and, for some factors, inconsistent.

Diabetes Diabetes increases cardiovascular morbidity and mortality 2- to 4-fold via multiple mechanisms, including prothrombotic alterations in blood coagulation. Unfavorable fibrin clot properties, with compact clot structure, have been observed in type 1 or type 2 diabetes since mid-1990s. Increased fibrinogen levels observed in diabetes correlate with the degree of hyperglycemia[21] and with fibrinogen glycation affecting fibrin polymerization and FXIII-mediated cross-linking.[22] Of note, impaired metabolic control and marked fluctuations in plasma glucose levels have been shown to be related to hypofibrinolysis associated with the formation of dense fibrin clots.[23]

Cigarette smoking Smoking unfavorably alters plasma fibrin clot structure and function. Acute cigarette smoking exposure has been demonstrated to be associated with more compact fibrin clots composed of thinner fibers compared with baseline and normal conditions.[24]

In chronic smokers, cigarette smoking leads to lower plasma fibrin clot permeability and longer clot lysis.[25] It is unknown whether smoking cessation restores normal fibrin clot properties.

Arterial hypertension Hypertension is a well-known cause of cardio- and cerebrovascular complications and numerous studies have demonstrated prothrombotic alterations detectable in circulating blood of patients with this disease. Very recently, it has been shown that essential arterial hypertension is also associated with reduced clot permeability, impaired clot lysability, and faster fibrin formation in plasma-based assays.[26] Effective antihypertensive treatment has been found to be associated with increased clot permeability and more efficient clot lysis following 6 months of therapy, and this effect correlated with reduction in systolic blood pressure.[26] Moreover, this small study suggests that improvement in fibrin clot parameters can be observed regardless of the class of hypertensive agents used.[26]

Hyperlipidemia Hyperlipidemia, especially hypercholesterolemia, has been shown to be inconsistently linked with altered plasma fibrin clot characteristics,[19] with much evidence indicating that elevated cholesterol levels cannot modify plasma clot properties. Recently, in hypertensive young and middle-aged patients we observed a significant negative correlation between total cholesterol and clot permeability,[26] indicating that at least in some populations unfavorable clot properties assessed in plasma might be to some extent associated with hypercholesterolemia.

Additional modifiers of fibrin clot properties in cardiovascular disease

Hyperhomocysteinemia Hyperhomocysteinemia defined as elevated homocysteine (Hcy) levels >15 μmol/l, has been reported to result in the formation of fibrin clots made of thinner and more tightly packed fibers and such clots are relatively resistant to plasmin.[27] It has been reported that 10 lysines in the D- and αC-domains of fibrinogen can be Nε-homocysteinylated by a highly reactive thioester, homocysteine thiolactone.[28] In up to 4% of the fibrinogen molecules Nε-homocysteinylation has been detected in cystathionine β-synthase-deficient patients.[29] Even in patients with advanced coronary atherosclerosis, elevated Hcy levels are associated with reduced clot porosity and enhanced lysis resistance.[30] However, no effect of Hcy on fibrin clot properties can be observed in the presence of diabetes or marked hypercholesterolemia.[30]

Lipoprotein(a) Lipoprotein(a) [Lp(a)], containing apo(a) domains homologous with the kringle domains IV and V in the plasminogen molecule, can compete with plasminogen for fibrin(ogen) binding sites, resulting in impaired clot lysis.[2] Elevated plasma Lp(a) levels have been found to correlate with reduced clot permeability, formation of thinner fibrin fibers, and impaired fibrinolysis both in apparently healthy subjects and patients with advanced coronary atherosclerosis,[31] in particular in the presence of small apo(a) isoformes.

Thrombin Thrombin formed during prothrombin activation by activated factor X in the prothrombinase complex modifies fibrin clot structure. Low prothrombin concentrations result in the formation of thicker fibers in fibrin clots characterized by increased permeability.[32] In the purified fibrinogen or plasma systems, clots produced with high thrombin concentrations are characterized by thin fibers that form a network of smaller pores.[8,19] Medications lowering thrombin formation or activity (e.g., vitamin K antagonists, heparins, fondaparinux, dabigatran, and rivaroxaban) improve fibrin clot phenotype, largely by increasing clot permeability.[33] It is worth noting that thrombin-induced alterations in fibrin formation are influenced by cellular procoagulant activity.[34] A denser fibrin network that is lysed more slowly is formed close to the cell surface of fibroblasts in the presence of the prothrombinase complex and fibrinogen or plasma.[34] Prothrombotic phenotype of the endothelium following exposure to inflammatory cytokines, most likely via TF expression, is associated with the formation of compact fibrin networks resistant to lysis.[35]

Oxidative stress Oxidative stress may alter fibrin clot properties via oxidation of fibrinogen and other proteins. Most in vitro experiments demonstrated that oxidation of fibrinogen impairs fibrin clot formation.[2,36] Carbonylation of proteins, including fibrinogen, particularly the Aα chain, has been demonstrated to be associated with higher rates of fibrin polymerization and thicker fibril formation.[37] Data on the association between oxidative stress markers and fibrin clot properties assessed in vivo are scarce. F_2-isoprostanes, which are produced upon nonenzymatic arachidonic acid peroxidation, used as a stable marker of oxidative stress measured in plasma or serum, have been shown to correlate with clot permeability and fibrinolysis in patients.[38]

Platelet activation Platelet activation, reflected by the release of a number of substances including proteins from α granules, has been shown to unfavorably alter clot properties, particularly at the sites of platelet aggregation.[6] For example, platelet factor 4 changes in vitro fibrin clot structure by the formation of an extremely compact fibrin network.[39] Polyphosphate, a polymer of inorganic phosphate secreted by activated platelets, has been demonstrated to alter

fibrin clot architecture and properties, including plasmin-mediated degradation.[40] Recently, platelet-derived pyrophosphate has been reported to counteract polyphosphate-induced changes in fibrin clot characteristics.[41] Fibrin clot architecture is affected not only by the substances released from platelets, but also by the sole presence of activated platelets. It has been shown that fibers originating from platelet aggregates are thinner and oriented radially.[42] Such clots are more stable and resistant to lysis.[42] In patients with chronic inflammatory states, elevated platelet count is associated inversely with clot permeability and positively with lysis time.[43] One might expect that classic platelet activation measures, first of all platelet aggregation stimulated by various agonists, are associated with altered clot properties. Indeed thromboelastographic parameters, such as clot strength, a measure of thrombin-induced fibrin and platelet interactions, correlate with adenosine diphosphate-induced platelet aggregation,[44] which might support the role of fibrin in prothrombotic phenotype observed in cardiovascular patients with high platelet activation.

Fibrin clot properties and cardiovascular medication Among medications used in therapy of patients with cardiovascular disease (apart from anticoagulant agents), 2 cardiovascular drugs of different pharmacokinetics and pharmacodynamics have been shown to improve fibrin clot phenotype in more than 1 study.

The first drug displaying fibrin altering properties is acetylsalicylic acid, aspirin, which in several models has been shown to increase clot permeability. A daily dose of 320 mg appears to produce a weaker effect compared with low-dose aspirin, which is most commonly used in Europe.[45] Aspirin-related increase in the fibrin clot pore size in stable coronary artery disease (CAD) has also been associated with enhanced clot lysability, which is likely the result of thicker fiber formation.[46] Aspirin alters fibrin clots by rendering them looser and their fibers thicker, and it reduces clot rigidity by 30% and enhances clot lysis, which has been confirmed using fibrinogen from healthy individuals receiving 150 mg of aspirin for 7 days.[47] Following exposure to aspirin acetyl groups can bind to the Aα chain of fibrinogen.[47] Acetylation of fibrinogen is the main mechanism that accounts for alterations in fibrin clot phenotype observed both in plasma and in purified systems. Moreover, at least in vitro aspirin may inhibit fibrinogen oxidation,[48] and in vivo aspirin at therapeutic doses can inhibit FXIII activation, thus affecting fibrin clot stability.[49]

Other popular medications altering fibrin clot phenotype are statins, which have been reported to produce a number of additional, so called pleiotropic, effects including antithrombotic properties.[50] A 4-week therapy with simvastatin, or atorvastatin (20–40 mg once daily) may increase fibrin gel permeability and accelerate clot fibrinolysis.[51] The magnitude of these effects induced by both statins was similar. Both fibrin clot measures were associated with statin-induced reduction in thrombin generation following vascular injury, most likely resulting from down-regulation of TF expression.[51] In subjects with low-density lipoprotein cholesterol below 3.4 mmol/l, simvastatin at a dose of 40 mg/d increases plasma fibrin clot permeability associated with faster clot lysis.[52] This effect was cholesterol-independent and correlated with reduction in C-reactive protein (CRP).[52] It remains to be established whether statins and aspirin may contribute to clinical benefits from their use.

Some other agents administered in cardiovascular disease have been suggested to improve clot phenotype. An angiotensin-converting enzyme inhibitor, quinapril (10 mg/d) given for 1 month, can increase clot permeability, independently of its antihypertensive effect.[51] Very recently, it has been demonstrated that polyunsaturated fatty acids (PUFAs), omega-3 acids, given at a dose of 1 g daily on top of recommended double antiplatelet therapy for 1 month can reduce fibrin clot permeability and increased lysability in patients with CAD undergoing elective angioplasty.[53] This effect is to some extent related to reduced thrombin generation following PUFAs administration.[53] Further studies are needed to confirm these observations.

Clot properties in atherosclerosis **Coronary artery disease** In 1992, the first report on fibrin clot characteristics in patients with advanced CAD has been published.[54] A major finding was that in such patients, plasma fibrin clots are less permeable (by 30%), indicating that fiber networks tend to become denser in this disease. Interestingly, fibrin clots with tightly packed, thin fibers and small pores are associated with the number and severity of coronary artery stenoses documented by angiography.[54] The clot phenotype was demonstrated in men with a history of myocardial infarction (MI), aged below 45 years.[55] Low clot permeability and prolonged lysis have been also observed in CAD in patients aged 60 years or more.[56] Compelling evidence indicates that fibrin is a consistent component of atherosclerotic plaques, which can promote their growth,[57] and thus fibrin may enhance cell proliferation participating in the progression of atherosclerosis.

Additional unfavorable clot features following MI have been reported by Collet et al.[58] who showed that fibrin clots obtained from 33 young individuals with prior MI had increased stiffness and number of shorter fibers, accompanied by impaired fibrinolysis compared with healthy controls. Patients aged below 50 years after a first MI, but not older individuals, had prolonged lysis time associated with the body mass index, blood pressure, and inflammatory state.[59] It appears that there is a link between genetic factors and fibrin clot characteristics in CAD. It has been reported that first-degree relatives of patients

who survived a first MI have similar, but milder alterations in the fibrin clot phenotype.[60]

Accumulating evidence suggests that thickness of fibrin fibers and the structure of their networks formed in vivo is a dynamic process associated with vital clinical variables in patients with acute MI. In March 2011, a French-American study showed that intracoronary thrombi aspirated from coronary arteries within the first 12 hours since symptom onset in ST elevation MI patients are rich in fibrin (56% vs. platelets, 16% of the content) and the amount of fibrin within the intracoronary thrombus is doubled every hour after the onset of chest pain.[61] Time of coronary ischemia has been found to be the only independent predictor of fibrin content in intracoronary thrombi.[61] Importantly, it has been elegantly demonstrated that platelet content in the thrombi and soluble CD40 ligand, a platelet activation marker, are inversely associated with fibrin content.[61]

Compared to stable CAD, acute MI patients display the tendency to form less permeable and lysable plasma fibrin clots that are composed of thicker fibers.[38] In contrast to stable angina, clot permeability and fibrinolysis in acute MI patients assessed within the first 12 hours since pain onset are correlated with oxidative stress and inflammation.[38] Another potent modulator of fibrin clot properties in acute MI is acute hyperglycemia observed in up to 50% of the patients. Elevated glucose in such clinical setting has been reported to prolong plasma fibrin clot lysis time without any association with clot permeability.[62] It is unknown whether ex vivo plasma fibrin clot properties are associated with fibrin content or characteristics in intravascular thrombi removed during invasive procedures, reflecting the in vivo situation. This issue appears of paramount importance from the pathophysiological point of view.

Fibrin clot properties have been shown to be implicated in 2 specific and life-threatening complications associated with invasive treatment of CAD. The first one is acute, subacute, and late stent thrombosis.,[63] and the other is the no-reflow phenomenon, i.e., the absence of a complete myocardial perfusion despite successful opening of the infarct-related artery.[64] Based on autopsy studies showing a lack of complete endothelialization and persistent fibrin thrombi as a primary substrate underlying stent thrombosis, we sought to investigate clot permeability and susceptibility to lysis in patients who survived such thrombotic event. A major finding was that a more tightly packed and less porous fibrin structure can be detected more commonly in patients with stent thrombosis.[63] These alterations might prolong the presence of fibrin in the lumen. Similarly, abnormal fibrin clot structure and function have been observed in patients with a history of the no-reflow phenomenon.[64]

Since several chronic diseases markedly increase the risk of MI, fibrin clot properties in such common disorders have been studied and abnormalities in clot phenotype assessed in plasma-based assays have been reported. In end-stage renal disease (ESRD), which is associated with substantially increased cardiovascular morbidity and mortality, it has been demonstrated that fibrin clots obtained from plasma of ESRD patients are less permeable and less susceptible to fibrinolysis than clots made from control plasma.[65,66] Importantly, clots made from plasma taken from patients who died of cardiovascular causes during a 3-year follow-up are less permeable and lysable than those from plasma of the remaining patients,[66] which indicates that fibrin clot phenotype may have a predictive value in terms of cardiovascular mortality. Growing evidence indicates that chronic inflammatory diseases via multiple mechanisms are associated with abnormal prothrombotic fibrin clot phenotype. Chronic obstructive pulmonary disease (COPD) is the best example of inflammatory disease consistently linked with 2- to 4-fold increased cardiovascular morbidity, which cannot be fully explained by smoking or other risk factors. Unfavorably altered clot permeability and lysis time in COPD patients were determined by the degree of stimulation of inflammation, as evidenced by their associations with CRP levels, which were more potent than those for fibrinogen.[67] Since CRP binds to fibrino(ogen),[68] it is likely that CRP alters fibrin network formation. CRP has been shown to inversely correlate with clot permeability and susceptibility to lysis both in healthy subjects and CAD patients.[38,56] Another example is rheumatoid arthritis (RA) associated with a 20-fold higher cardiovascular risk compared with the general population. Patients with RA had lower clot permeability, faster clot formation, higher maximum clot absorbancy indicating thicker fibrin fibers, and prolonged fibrinolysis time than controls.[43]

Ischemic stroke Cerebrovascular ischemic events have been demonstrated to form more compact (by 20%) fibrin clots compared with controls, with no differences in their lysability.[69] Fibrin properties have been shown to be correlated with stroke severity, but not with post-stroke mortality during follow-up.[69] What is important, ischemic stroke of unknown origin, representing about one quarter of all cases, is characterized by prothrombotic plasma fibrin clot phenotype. It has been shown that patients with previous cryptogenic stroke tend to form dense plasma fibrin clots resistant to lysis.[70] Similarly to acute MI patients,[38] acute ischemic stroke within the first 72 hours of symptom onset has been demonstrated to be associated with reduced clot permeability and susceptibility to lysis.[71] It is worth noting that coexistence of acute ischemic stroke and CAD prolongs clot lysis as compared with the findings observed in stroke not associated with concomitant CAD.[71] However, links between clot phenotype and clinical outcomes in acute stroke are inconsistent. Only fibrin clot compaction has been

demonstrated to correlate with neurological deficit both on admission and at discharge.[71] Interestingly, lower fibrin clot permeability and reduced fibrinolysis observed in the acute phase of ischemic stroke do not change after 60 days from the event,[72] suggesting the hypofibrinolysis is a persistent characteristic of ischemic stroke. Taken together, ischemic stroke is associated with alterations in fibrin clot characteristics, suggesting mechanistic similarities in thrombotic arterial episodes in various vascular locations. It is probable, however, that not all types of ischemic stroke are characterized by the same magnitude of abnormalities in fibrin clot characteristics both in the acute phase and following this event. The hypothesis of weaker associations between lacunar stroke and abnormal fibrin clot phenotype merits investigation.

Peripheral arterial disease Peripheral atherosclerotic occlusive disease (PAD), resulting from progressive narrowing or occlusion of the major arteries in the lower limbs caused by atherosclerosis, affects as many as up to 30% of elderly patients, and approximately two-thirds of patients with the disease are asymptomatic.[73] PAD is associated with other manifestations of cardiovascular disease, as evidenced by the findings showing that the 5-year incidence of PAD in elderly men amounts to 29% for subjects following cerebrovascular events and 25% for patients with CAD.[73]

In middle-aged and elderly PAD patients, it has been shown that plasma fibrin clots contain smaller pores, are composed of thicker fibers, are formed more rapidly, and are lysed at a reduced rate, compared with those made from plasma obtained from controls.[74] These findings indicate that patients with PAD share several common clot characteristics with subjects suffering from CAD. Importantly, unfavorable fibrin clot phenotype, reflected by the tendency to form compact and poorly lysable clots, might predispose to occlusive vascular complications and herald worse prognosis.[74] In 34 relatively young patients with mild to moderate PAD, plasma fibrin clots have also been reported to be poorly permeable, rigid, and resistant to lysis.[75] The most pronounced and typical feature of PAD patients appeared to be increased fiber thickness on turbidimetry.[75] In 2009, Guimaraes et al.[76] showed that hypofibrinolysis was associated with a 2.3-fold higher odds ratio of PAD diagnosed at a young age. Of note, first-degree relatives of PAD patients had similarly altered clot characteristics as the patients,[77] indicating the importance of yet unidentified genetic factors that might be implicated in the pathogenesis of PAD, particularly in younger age groups. Collectively, current evidence points to the role of unfavorably altered clot characteristics in the development and progression of PAD.

Conclusions Atherothrombotic vascular disease remains a major cause of morbidity and mortality worldwide. Several lines of evidence, derived largely from small case-control studies, support the concept that particular prothrombotic fibrin clot phenotype contributes not only to acute thromboembolic complications of this disease, but also may be implicated in the progression of atherothrombosis. It is likely that abnormal fibrin clot phenotype characterizes all patients with atherosclerotic vascular disease. Such clot phenotype that can be evaluated in clinical settings might be a novel risk factor for atherothrombosis. Altered clot architecture in patients at cardiovascular risk may be attributed to a number of factors, including those associated with classic and novel risk factors, e.g., elevated levels of Hcy and Lp(a). Most fibrin alterations in this context can be summarized as follows: fibrin clots formed by tightly packed thin fibers with small pores are resistant to lysis. From a clinical point of view, it remains to be established to what extent changes in fibrin clot properties are associated with clinical endpoints and whether fibrin network modulation by, for example, statins is clinically relevant. Large prospective follow-up studies are needed to address the pivotal issue as to whether fibrin clot parameters might help identify subjects at risk of MI or stroke in the future, especially among younger individuals without traditional risk factors. Undoubtedly, research on the links between coagulation, inflammation, and atherosclerosis in the context of altered fibrin clot properties is still a challenge and several questions await reliable answers to optimize risk assessment in at-risk subjects.

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ARTYKUŁ POGLĄDOWY

Nabyta dysfibrinogenemia w chorobach naczyń na podłożu miażdżycy

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SŁOWA KLUCZOWE

choroba wieńcowa, fibrynoliza, miażdżyca, skrzep fibrynowy, udar mózgu

STRESZCZENIE

Nabyte jakościowe nieprawidłowości w cząsteczkach fibrynogenu, zwane nabytą dysfibrynogenemią, wykazano w wielu chorobach przeważnie związanych ze stanem prozakrzepowym, w tym w szpiczaku mnogim i chorobach wątroby. Fibryna występuje w dużych ilościach w blaszkach miażdżycowych. Zmienione właściwości osoczowego skrzepu fibrynowego, zwykle odzwierciedlone przez zmniejszoną przepuszczalność skrzepu i upośledzoną fibrynolizę, stwierdzono u chorych z świeżym lub przebytym zawałem serca, z niedokrwiennym udarem mózgu oraz z miażdżycą zarostową tętnic kończyn dolnych. Ponadto prozakrzepowy fenotyp skrzepu fibrynowego obserwowano u pacjentów, u których wcześniej wystąpiło zjawisko braku powrotu przepływu (*no-reflow phenomenon*) lub zakrzepica w stencie. Coraz więcej danych wskazuje, że nabyta dysfibrynogenemia przyczynia się do postępu choroby naczyń na podłożu miażdżycy i występowania jej zakrzepowych powikłań. Ten artykuł podsumowuje aktualny stan wiedzy na temat związków między prozakrzepowym fenotypem skrzepu fibrynowego a chorobą naczyń na podłożu miażdżycy oraz opisuje szerokie spektrum czynników ryzyka sercowo-naczyniowego jako czynników modyfikujących cechy charakteryzujące sieć fibrynową.

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