

Is Fibrinogen a Marker of Inflammation in Coronary Artery Disease?

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Evidence suggests that thrombosis, endothelial dysfunction and inflammation are strongly associated with coronary artery disease (CAD). Inflammation plays a crucial role in characterizing the formation of atheromatous plaque, as well as its progress.¹⁻⁶ The secretion of proinflammatory cytokines from the vascular endothelium as well as from macrophages induces the production of inflammatory molecules that are measured in the circulation, such as C-reactive protein (CRP), serum amyloid A and fibrinogen.^{1,5,7}

Several studies have focused on fibrinogen, demonstrating a strong association with the presence of CAD, while others failed to show a significant association. Although most of the available data favor the involvement of fibrinogen in CAD, there is still evidence against this hypothesis.

Link between inflammation and CAD: a role for fibrinogen

Fibrinogen and its metabolites may lead to endothelial dysfunction through various mechanisms.⁸ Several atherosclerotic lesions contain large amounts of fibrin, either in the form of wall thrombus in the intact surface of the plaque or scattered diffusely all over the plaque. This phenomenon is associated with a decrease in fibrinolytic activity and plasminogen concentrations, states that are observed

in CAD.⁹ It has been found that fibrin (intima) triggers cell proliferation, contributing to cell migration, and bonds fibronectin, which triggers cell migration and adhesion.¹⁰ Fibrinogen and products of its decomposition mediate the transportation of adhesion molecules in the surface of endothelium and their further migration to the intima.¹¹ The decomposition products located in the inner layer can trigger mitogenesis and synthesis of collagen, attract leukocytes, and enhance permeability as well as vascular tone. In advanced atherosclerotic plaques fibrin participates in the close linkage of low-density lipoprotein (LDL) and lipid accumulation, leading to the creation of the lipid nucleus of atherosclerotic lesions.^{9,12}

In addition, proinflammatory cytokines, such as interleukin 6 (IL-6) and tumor necrosis factor alpha (TNF- α), are produced from the vasculature, adipose tissue and myocardium; these increase the synthesis of nitric oxide (NO) and favor leukocyte migration in the sub-endothelial space. These cytokines also have a further regulatory role, inducing liver synthesis of acute phase proteins, such as fibrinogen, and consequently inflammatory and prothrombotic reactions.¹³ Thus, fibrinogen participates in the formation of atherosclerotic plaque during the first stages of CAD, suggesting that it is a causative factor rather than a result.

Evidence supporting the association between fibrinogen and CAD

Early evidence from previous decades pointed to a strong association between fibrinogen levels and CAD manifestation (Table 1).

The Gothenburg Study¹⁴ reported that plasma fibrinogen levels represent an independent risk factor for myocardial infarction (MI) and stroke in univariate analysis. Similarly, the Framingham study^{15,16} demonstrated that the risk for MI and stroke increased progressively along with fibrinogen levels. The effect of fibrinogen levels on cardiovascular risk was even greater in young individuals and was similar to the effect of known risk factors such as hypertension, diabetes mellitus, and smoking. Another large epidemiological study showed that fibrinogen was not only a strong and independent risk factor for MI and sudden cardiac death in patients with pre-existing CAD, but also had a greater predictive value for future coronary events, compared to von Willebrand factor (vWF) antigen and tissue plasminogen activator antigen.¹⁷ Novel data from the EPIC-Norfolk study¹⁸ showed prospectively that fibrinogen levels were significantly higher in patients presenting with fatal or non-fatal coronary heart disease, than in those remaining free of any cardiovascular disease during follow up, while Acevedo et al¹⁹ reported that fibrinogen was directly associated with the presence of MI and was revealed to be an independent short-term predictor of mortality. One multivariate analysis found an independent association between dual parental and sibling history of MI and plasma fibrinogen levels,²⁰ implying that plasma fibrinogen levels may be an inheritable risk factor for CAD in subjects with a strong family history of MI. Interestingly, increased levels of fibrinogen have been correlated with adverse cardiac events after intracoronary stenting, suggesting a potential role of fibrinogen levels in the outcomes following percutaneous coronary interventions.²¹

Despite the data supporting a role of fibrinogen as a marker of CAD and its manifestations, several studies have investigated the role of fibrinogen as a risk factor-mediator of CAD. Recently, Shojaie et al²² and Pineda et al²³ introduced high levels of fibrinogen as a risk factor for premature CAD in subjects <55 years. A role in subclinical atherosclerosis has been also attributed to this acute phase protein, as higher levels of fibrinogen during young adulthood were positively associated with prevalence of coronary artery calcification and increased carotid intimal-medial thickness in middle age, while

the magnitude of the association decreased with aging.²⁴ Similarly, fibrinogen and CRP levels were strong predictors of subclinical atherosclerosis (associated with an extension of carotid atherosclerosis) in hypertensive postmenopausal women.²⁵ Supporting data had previously shown that fibrinogen is involved in the subclinical phase of extracoronary and coronary atherosclerosis and may add to the atherogenic effect of hyperlipidemia.²⁶ Moreover, fibrinogen levels have been found to be independently related to cardiovascular mortality, extent,²⁷ as well as the severity of disease.²⁸ Moreover, fibrinogen levels were significantly higher in patients presenting with unstable than in those with stable angina, suggesting a role for fibrinogen in the pathophysiology of acute coronary syndromes.²⁸

Conflicting data have been reported by other studies and show no association between fibrinogen levels and CAD (Table 1).²⁹⁻³⁵ However, these results appear surprising, given that the role of fibrinogen in the pathogenesis of CAD has been well evaluated by the majority of the available studies.

The genetic view

There are several gene polymorphisms of fibrinogen chains, recognized during the last decade, which seem to affect fibrinogen levels and consequently the presence of CAD (Table 2).

Green et al³⁶ showed a significant interaction between smoking status and genotype in determining plasma fibrinogen levels in healthy individuals, providing an independent confirmation of the association between beta(b)-fibrinogen G455A genotype and plasma fibrinogen levels. Similarly, it has been observed that polymorphisms G455A and G845A of the b-chain gene of fibrinogen significantly affect the concentration of the latter.³⁷ In particular, the -455G/A polymorphism is one of the most important genetic variables associated with elevated fibrinogen levels in both genders in the general population.³⁸ Interestingly, in patients with diabetes mellitus type 2 there was a strong relationship between the -455G/A b-fibrinogen gene polymorphism and the development of CAD.³⁹ Additional data have also been reported regarding the same gene polymorphism.⁴⁰ In patients with stable CAD and healthy individuals, the G455A polymorphism increased fibrinogen levels significantly in the wild genotype in patients with CAD; however, no significant effect was observed among healthy individuals.

In addition, fibrinogen levels have been affected

Table 1. Studies focusing on the association between fibrinogen and coronary artery disease.

Study	Population	No.	Years	Comments on fibrinogen and CAD	Level of significance
Wilhelmsen et al ¹⁴	General population	792	13.5	Fibrinogen levels strongly associated with stroke and MI.	-
Kannel et al ¹⁵	Healthy	1315	12	Fibrinogen is a predictor of CVD.	-
Kannel et al ¹⁶	Healthy	1314	16	Diabetics presented with higher fibrinogen levels.	-
Thompson et al ¹⁷	Angina pectoris	3043	2	Increased incidence of MI or sudden death with higher fibrinogen levels.	p=0.01
Rana et al ¹⁸	Healthy	2550	6	Fibrinogen levels were significantly higher in subjects with CAD. High c statistic for FRS plus fibrinogen.	p<0.001
Acevedo et al ¹⁹	CAD vs. Healthy	2126	-	Fibrinogen directly associated with MI. Independent predictor of mortality.	p=0.001 (HR=1.81, p<0.001)
Robinson et al ²⁰	FH of MI vs. Healthy	170	-	Subjects with a dual parental and sibling history of MI had higher fibrinogen levels.	p<0.05
Germing et al ²¹	CAD	228	-	Significantly higher level of fibrinogen in the group with adverse cardiac events compared with controls.	p=0.028
Shojaie et al ²²	MI vs. Healthy	66	2	Plasma fibrinogen levels were significantly higher in subjects with MI than in the control group.	p=0.036
Pineda et al ²³	MI vs. Healthy	237	-	MI patients had significantly higher fibrinogen levels. In a multivariate analysis, fibrinogen levels were independently associated with presence of MI.	p=0.006 p=0.038
Green et al ²⁴	General population	2832	13	Fibrinogen was positively associated with greater CAC prevalence and increased CIMT. After adjustment positive relationships between fibrinogen and CAC prevalence remained for the total cohort longitudinally, but not cross-sectionally.	p<0.001 p=0.037 and p=0.147
Rizzo et al ²⁵	HT (PM) women	127	5	Fibrinogen levels were associated with the extension of carotid atherosclerosis. Elevated fibrinogen was an independent predictor of subclinical atherosclerosis.	p<0.0001 p=0.0298
Levenson et al ²⁶	Hypercholesterolemic	693	-	Fibrinogen levels were higher in the diseased (1, 2, 3, 4) sites than the non diseased sites.	p<0.05, p<0.01, p<0.001 and p<0.001, respectively
Espinola-Klein et al ²⁷	Undergoing coronary angiography	719	6.5	Greater mortality in those with high fibrinogen levels. Predictive value of fibrinogen for mortality.	p<0.0001 HRR: (95% CI) 2.2 (1.2–3.9), p<0.01
Gil et al ²⁸	ACS vs. stable angina	101	-	Plasma fibrinogen levels were significantly higher in patients presenting with unstable angina. Significant correlation between fibrinogen and troponin I levels in unstable patients.	p=0.002 p=0.0015
Cremer et al ^{29,30}	Healthy	6002	5	When adjusted for LDL, there was no significant association between plasma fibrinogen CAD without MI.	p=NS
Lawlor et al ³¹	CAD vs. Healthy	721	-	Adjustment for all potential confounding factors attenuated the association between fibrinogen and CAD.	OR: 1.29 (1.12, 1.49) to 1.09 (0.93, 1.28).

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Hartmann et al ³²	LMC	60	0.75-2.25	Association between fibrinogen and changes in plaque-plus-media area. Patients with plaque progression had higher fibrinogen levels. Multivariate analysis showed no independent association between fibrinogen and plaque progression. Multivariate analysis showed that fibrinogen levels were independently associated with adverse cardiovascular events.	p<0.0001 p=0.019 p=NS p=0.018
Cavusoglu et al ³³	Undergoing coronary angiography	219	-	Fibrinogen was significantly higher in patients with CAD. No difference was found in fibrinogen levels among the patients with single, double or triple vessel disease.	p<0.01 p=NS
Tousoulis et al ³⁴	MI	30		Fibrinogen levels are not associated with the presence or absence of Q waves.	p=NS
Sjoland et al ³⁵	CABG	729	10	Patients with high levels had increased mortality compared to fibrinogen levels below median. After adjustment for specific factors pre-operative fibrinogen was not an independent predictor of long-term mortality.	p=0.0005 p=NS

ACS – acute coronary syndromes; CABG – coronary artery bypass grafting; CAC – coronary artery calcification; CAD – coronary artery disease; CI – confidence interval; CIMT – carotid intima-media thickness; CVD – cardiovascular disease; FH – family history; FRS – Framingham risk score; HR – hazard ratio; HRR – hazard risk ratios; HT – hypertensive; LDL – low-density lipoprotein; LMC – left main coronary; MI – myocardial infarction; NS – non significant; OR – odds ratio; PM – postmenopausal.

Table 2. Genetic polymorphisms associated with fibrinogen levels and CAD.

Study	Population	No.	Gene polymorphisms	Effects on fibrinogen and CAD	Level of significance
Green et al ³⁶	MI and Healthy	209	G455A (b-chain)	Genotype was significantly associated with (adjusted) plasma fibrinogen level in controls. Mean fibrinogen levels (smokers) were significantly higher in G/A-455 and A/A-455 individuals compared with G/G-455 homozygotes.	p=0.003 p=0.001
van't Hooft et al ³⁷	Healthy	210	G455A, G854A (b-chain)	The influences of the G455A and G854A polymorphisms on the plasma fibrinogen concentration were about equally strong when the effects of age and smoking habits were considered. For the G455A/G854A the results showed significant increase of fibrinogen levels in GG/AA, AA/GG, GA/GA.	G455A: p<0.05 (AA, AG for the common allele) G854A: p<0.05 (AA for the common allele) Overall p<0.0001 for both in the ANCOVA p<0.05 for all compared with the GG/GG

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Tybjaerg-Hansen et al ³⁸	General population	9127	G455A (b-chain)	The A allele was associated with elevated plasma fibrinogen levels in both genders. An increase of 1 SD in plasma fibrinogen increased the odds ratio for ischemic heart disease by approximately 20%. Fibrinogen is not a predictor of ischemic heart disease.	p<0.001 p<0.01 for women, p<0.005 for men p=NS
Carter et al ³⁹	NIDDM with CAD or no CAD	187	G455A (b-chain)	Significant difference between subjects with CAD and those without CAD. Increased risk for CAD in individuals homozygous for the G allele vs. A allele. Increased risk for CAD with an increase of 1 g/l in fibrinogen levels.	p=0.004 OR: 1.77 (1.08-2.90), p=0.03 OR: 1.60 (1.00-2.60), p=0.05
Papageorgiou et al ⁴⁰	CAD vs. Healthy	243	G455A (b-chain)	Fibrinogen levels were much higher in CAD than controls for the G455A polymorphism. (In CAD) AA patients had significantly higher levels of fibrinogen only than GA patients. (In controls) there were no differences across the genotypes. AA and GG patients with CAD and significant higher levels of fibrinogen than AA and GG controls. AG controls had significant higher fibrinogen levels than AG CAD. CAD patients with AA and GG genotypes had significantly higher levels of D-dimers than controls, while there was no difference for AG genotype.	p<0.001 p<0.05 p=NS p<0.01 and p<0.05 p<0.05 p<0.05, p<0.05, p=NS
Tousoulis et al ⁴¹	CAD or no CAD Hypertensives	412	A1675G (AT2R)	The G allele was associated with decreased risk of CAD among hypertensives and less aggressive angiographic CAD. The G allele was associated with lower fibrinogen vs. A allele. The effect of A1675G on fibrinogen was driven by its effect among hypertensives (Avs.G) whereas it had no effect among non-hypertensives.	OR: 95% CI: 0.4 (0.2–0.9), p<0.01 p<0.001
Bockholdt et al ⁴²	CAD vs. Healthy	2478	Val34Leu (factor XIII)	Significant interaction between the Val34Leu variant and fibrinogen levels for the risk of future CAD. Among people in the lowest tertile of fibrinogen concentrations, LeuLeu carriers had increased risk for CAD compared to wild-type individuals. Among those in the highest fibrinogen tertile, LeuLeu carriers had a lower risk than wild-type individuals.	p=0.004 OR: 2.88 (CI: 1.24-6.74), p=0.003 OR: 0.47 (CI: 0.18-1.17), p=0.1
Mannila et al ⁴³	MI	774	T9340C (γ-chain) G2224A (α-chain) G1038A (b-chain)	Significant interaction between the b-chain G1038A and case-control status on plasma fibrinogen gamma. The T9340C and G2224A appeared to interact on the plasma fibrinogen gamma concentration. The risk of MI was associated with an increased fibrinogen gamma concentration and remained significant after controlling for the T9340C and G2224A. The plasma fibrinogen gamma concentration was involved in a high-order interaction with total plasma fibrinogen and T9340C and G2224A polymorphisms, associated with a further increased risk of MI. An elevated plasma fibrinogen gamma concentration was independent predictor of MI.	p=0.02 p<0.001 OR: (95% CI): 1.34 (1.16, 1.55), p<0.001 OR: (95% CI): 3.22 (2.35, 4.39), p<0.001 OR: (95% CI): 1.24 (1.01, 1.52), p=0.04

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Sun et al ⁴⁴	CAD	1254	C148T, G854A (b-chain)	Fibrinogen levels were related to CAD. Frequencies of fibrinogen C148T G854A alleles, gene and genotypes between the two groups were similar (no association with CAD).	p<0.05 p=NS
Doggen et al ⁴⁵	MI vs. Healthy	1206	G455A, BclI (b-chain) TaqI (a-chain)	The TaqI, HaeIII and BclI polymorphisms in the fibrinogen gene were not associated with MI.	p=NS for all comparisons
Blake et al ⁴⁶	MI, stroke, VT vs. Healthy	1502	C148T (b-chain)	Frequency of the T allele was similar among men who had MI, stroke or VT compared with those with no cardiovascular events. No evidence of association between the T allele and MI or VT.	p=NS for all comparisons RR: 1.06; 95% CI 0.82-1.36), stroke (0.87, 0.63-1.21) and (0.75; 0.51-1.08)
Leander et al ⁴⁷	MI vs. General population	2708	G455A (b-chain)	Significant higher levels of fibrinogen in men with MI. The -455A allele was not associated with an increased risk of MI. There were no strong indications of synergistic interaction between the G-455A polymorphism and any of the environmental exposures considered.	p<0.05 p=NS p=NS
Chen et al ⁴⁸	CAD vs. Healthy	2722, 2104	C148T, G455A (b-chain)	No increased risk of CAD in -148T allele carriers compared to the -148C/C wild-type homozygotes Susceptibility to CAD in -455A allele carriers compared to the -455G/G wild-type homozygotes	OR: 1.31; 95%CI: 0.94-1.84, p=0.11 OR: 1.75; 95%CI: 1.24-2.46, p=0.001
Kardys et al ⁴⁹	CAD vs. Healthy	6400	G58A, G1374A, T1526C, Thr312Ala (a-chain) G4288A, G6326A, T7792C (γ-chain)	Fibrinogen γ-chain and a-chain gene haplotypes were not associated with coronary events, coronary atherosclerosis or extracoronary atherosclerosis.	p=NS for all

A – adenine; a-chain – alpha chain; ANCOVA – analysis of covariance; AT2R – angiotensin type 2 receptor; b-chain – beta chain; γ-chain – gamma chain; C – cytosine; CAD – coronary artery disease; CI – confidence interval; CRP – C-reactive protein; FMD – flow-mediated dilatation; G – guanine; Leu – leucine; MI – myocardial infarction; NIDDM – non-insulin dependent diabetes mellitus; NS – non significant; OR – odds ratio; RR – relative risk; T – thymine; Val – valine; VT – venous thromboembolism.

by other gene polymorphisms, such as A1675G on angiotensin type 2 receptor gene, which was found to affect cardiovascular risk and the severity of atherosclerosis by modifying systemic inflammation (such as fibrinogen), especially in hypertensive males.⁴¹ A significant gene-covariate interaction has also been reported between the factor XIII (fXIII) Val34Leu variant and fibrinogen levels.⁴² Importantly, Manilla et al⁴³ demonstrated that plasma fibrinogen gamma' concentration influences the risk of MI, and this association seems to be enhanced by the presence of an elevated total plasma fibrinogen concentration and the fibrinogen gamma chain -9340T and fibrinogen alpha chain -2224G alleles.

Nevertheless, the results are controversial and some studies have failed to point out such correlations between specific gene polymorphisms and plasma fibrinogen levels or CAD development. Sun et al⁴⁴ found that fibrinogen levels were related to CAD. However, fibrinogen C148T and G854A polymorphisms were not associated with CAD. Despite the fact that a common mutation (G-455-->A) in the beta-fibrinogen promoter was an independent predictor of plasma fibrinogen, it was not a predictor of ischemic heart disease.³⁸ Moreover, a significant number of studies failed to ascertain the association between polymorphisms in the fibrinogen gene and cardiovascular risk.⁴⁵⁻⁴⁷ Novel data reported the absence of an association between the b-chain fibrinogen gene -148C/T polymorphism, while there is susceptibility to the risk of CAD for the G455A polymorphism.⁴⁸ Similarly, Kardys et al⁴⁹ showed that specific fibrinogen gene haplotypes were not associated with coronary events.

Conclusions

Fibrinogen represents an inflammatory marker that appears to be implicated in the pathophysiology and prognosis of CAD. Its presence contributes to the formation of atheromatous plaque, while it contributes to the development of acute coronary syndromes via its interaction with other inflammatory substances, endothelium and thrombotic molecules. The vast majority of studies focusing on the association between fibrinogen levels and CAD, even gene-related, demonstrated a positive correlation, while others failed to show any correlation. In addition, several gene polymorphisms of fibrinogen chains are associated with cardiovascular events and fibrinogen levels. Although fibrinogen is used widely in clinical practice and epidemiological studies have evaluated its role in CAD,

there are still aspects that need further investigation. Thus, many more large scale studies are required to evaluate the association of fibrinogen with advanced atherosclerosis.

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