Inherited thrombophilia and the risk of myocardial infarction: current evidence and uncertainties

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ABSTRACT
Atherothrombotic diseases (ATEs) and venous thromboembolism (VTE) have been traditionally considered to have a distinct pathogenesis. Today, a growing body of evidence on the pathophysiology of thrombus formation has convincingly proved that they share more mutual risk factors than previously recognized. It has been shown in a number of case-control studies that there is a significant risk for a subsequent cardiovascular disease after VTE, although this risk is low or at most moderate. In the past 2 decades, the role of each inherited risk factor for VTE in relation to ATE has been intensively studied. Unfortunately, a large body of contradictory findings has been published that hinders consensus and transformation of knowledge into clinical practice. Complicated gene-gene interactions, small sample sizes, heterogeneous genetic and environmental patient backgrounds, confounding factors, and varied methodological designs may have contributed to opposing findings. In the case of rare thrombophilias, conclusions must be summarized based on case reports or case series, as only few case-control and cohort studies are available. In this review we focus on available evidence and controversies regarding the relationship between the classic inherited VTE risk factors (factor V Leiden, prothrombin 20210A, deficiencies of antithrombin, protein C, and protein S) and the risk of myocardial infarction (MI). We conclude that the risk of MI in patients with common inherited thrombophilia is generally modest. However, in patients with deficiencies of antithrombin, protein C, or protein S, the risk of MI or other ATEs is not negligible. A personalized clinical approach is suggested when testing for inherited thrombophilia in a patient with MI.

KEY WORDS
acute myocardial infarction, factor V Leiden, protein C, protein S, thrombophilia

Introduction
Thrombosis is a common complex disease including atherothrombotic diseases (ATEs) and venous thromboembolism (VTE), which are highly frequent and are major contributors to morbidity and mortality in all developed countries. Clinically significant arterial thrombosis most typically occurs in the form of myocardial infarction (MI), ischemic stroke (IS), or peripheral arterial occlusive disease, while venous thromboses include deep vein thrombosis and pulmonary embolism. The term “thrombophilia” was first used by Nygaard and Brown in 1937, when they described 5 case reports with sudden occlusion of large arteries, sometimes with coexistent VTE, and linked the events to hypercoagulability of the patients’ plasma. Some decades later, research on thrombophilia expanded by investigating families with frequent or unusual events of VTE. Today, due to this field of extensive research, the term thrombophilia is used to describe a hemostasis disorder leading to an elevated risk of developing VTE that can be inherited, acquired, or both. The well-established factors of inherited thrombophilia are as follows: different loss-of-function mutations of the inhibitors of plasma coagulation that cause deficiencies of antithrombin (AT), protein C (PC), and protein
S (PS), gain-of-function mutations known as factor V Leiden (FVL) that result in resistance to activated PC (APC), and the guanine to adenine mutation at nucleotide 20210 in the 3’ untranslated region of prothrombin gene (FII20210A). Further hereditary conditions, which may contribute to thrombotic phenotype are non-O blood group, elevated factor VIII, IX, and XI levels, certain types of inherited fibrinogen disorders, and hyperhomocysteinemia. However, some of these latter risk factors are only partly inherited, for example, heritability of hyperhomocysteinemia is low, being rather related to several acquired conditions, predominantly to deficiencies of vitamin B₉ and folic acid. The level of factor VIII is in part determined genetically, but it is highly influenced by different clinical conditions (eg, acute phase reactions, VTE itself). Moreover, the cutoff value for elevated factor VIII is a matter of debate. Elevated levels of other clotting factors are rarely considered as the cause of thrombophilia, and therefore are not investigated routinely.

The discovery of hereditary thrombophilia risk factors raised considerable attention worldwide that initially led to a rising tendency to test for their presence in VTE. Unfortunately, widespread testing in nonselected patients, using various methodologies suffering from preanalytical and analytical issues, and unanswered questions regarding the clinical implications of the results often left treating clinicians disillusioned by the usefulness of testing. Up-to-date indications for thrombophilia testing in patients with VTE have been summarized in various guidelines and reviews; however, validated guidance is still lacking on whom and what to test. As no single well-standardized and widely accepted method exists for heritable thrombophilia screening, a list of investigations has to be performed in a patient suspected of thrombophilia. Thrombophilia screening usually includes tests for APC resistance or FVL, FII20210A, as well as assays for AT, PC, and PS. Some guidelines also recommend testing for elevated levels of FVIII and other clotting factors, members of the fibrinolytic system, for example, plasminogen activator inhibitor 1 (PAI-1), together with the 4G/5G polymorphism in the PAI-1 promoter; however, they have not been conclusively associated with VTE risk or require further validation.

Thrombophilia testing in ATE is rather controversial. Most of the guidelines do not recommend inherited thrombophilia testing in the case of arterial events, owing to the lack of evidence for their role in the pathogenesis of the disease and lack of justification regarding altered or intensified secondary prevention. However, a more recent concept of atherothrombosis suggests that arterial and venous thromboses have more in common than previously recognized. Although ATE and VTE have been traditionally considered to have distinct pathomechanisms and risk factors, today, a growing body of evidence on the pathophysiology of thrombus formation has convincingly questioned this dichotomy. Recent studies on the structure of arterial and venous thrombi have led us to the conclusion that although the environments in which these clots form and propagate do not overlap, the composition of these thrombi have many shared features. In studies when arterial, intracoronary thrombi were extracted by thromboaspiration from patients after MI, fibrin was found to be the main component of the thrombus, followed by lower platelet and red blood cell counts, and much fewer white cells. Notably, a microscopic analysis of venous thrombi revealed that they are also mostly composed of fibrin and entrapped red cells, with fewer platelets and leukocytes randomly dispersed, or concentrated in the caudal segment of these thrombi. Today, it is becoming clear that both arterial and venous thrombi are composed of a complex fibrin network and contain the same blood-borne cells, and it is only the relative content of these elements that may differ. It has been shown that genetic and environmental factors affect clot formation, structure, and stability. Based on common aspects in the pathophysiology of venous and arterial clot formation, it is plausible to think that in the presence of inherited or acquired risk factors of VTE, arterial clots might form more readily as well.

The concept of increased risk of subsequent arterial events in patients with VTE has been a matter of debate and has been tested in a number of case-control studies. A systematic review and meta-analysis of 17 studies showed that the risk of arterial cardiovascular events appeared to be higher in patients with unprovoked VTE as compared with controls (incidence rate ratio, 1.87; 95% CI, 1.32–2.65) and than in patients with provoked VTE (incidence rate ratio, 1.86; 95% CI, 1.19–2.89). In a large prospective cohort study including 4480 patients with VTE, it was found that patients have 2.2-fold higher risk of subsequent cardiovascular disease as compared with controls (95% CI, 1.2–3.8). After confounders (age, sex, body mass index, smoking, malignancy, chronic disease, genetic thrombophilia, procoagulant markers) were also considered, the risk estimate was no longer increased in patients as compared with controls. These results demonstrate that there is a risk for a subsequent cardiovascular disease after VTE, although this risk is low or at best moderate, and that the relationship between VTE and subsequent cardiovascular disease is not causal but can be explained by common etiologic factors, including that of inherited thrombophilia.

The contributing role of each particular inherited risk factor for VTE to arterial thrombotic
events has been studied intensively in the past 2 decades. In this review we focus on available evidence and controversies regarding the relationship between the classic inherited VTE risk factors (FVL, FII20210A, deficiencies of AT, PC, and PS) and the risk of MI.

**Factor V Leiden and the risk of myocardial infarction** Overview of factor V Leiden mutation

The first report on APC resistance in 1993 by Dahlbäck et al., followed by the identification of the responsible genetic defect in factor V (FV G1691A) in 1994, opened a new chapter in the history of thrombophilia research. The active form of FV acts as a cofactor to activated factor X in the process of converting prothrombin to thrombin. Factor V becomes activated by thrombin and its active form is susceptible to cleavage and inactivation by APC. In the presence of FVL mutation, an amino acid change takes place at position 506 of the mature protein (p.Arg506Gln, according to traditional nomenclature). As a result, the APC cleavage site is altered, rendering FVL resistant to proteolytic inactivation, resulting in a longer half-life and increased generation of thrombin.

The mutation of FVL is the most common thrombotic risk factor in the world, although it shows an uneven geographic distribution ranging from being practically absent in native Africans, Asians, Americans, or Australians, to being relatively frequent in populations of European origin, where allele frequency is generally between 3% to 7%, but may be as high as 10% or more in some populations. The relative risk of venous thrombosis associated with FVL has been clearly established (3- to 7-fold risk increase for homozygosity as compared to wild type individuals); however, the role of FVL as a risk factor for arterial events, and particularly for MI, remains controversial.

**Studies on the role of factor V Leiden mutation in the development of myocardial infarction**

Despite numerous studies and considerable efforts to understand the association of FVL with MI risk, conflicting results have been published. As of January 2019, a comprehensive PubMed search of the literature on this topic from the past 20 years resulted in more than 100 original publications. Unfortunately, no clear message emerges from these papers. Small sample sizes, heterogeneous genetic and environmental backgrounds, complicated gene–gene interactions, confounding factors, and varied methodological designs may have contributed to the opposing findings that were published.

Six meta-analyses were conducted on the subject (Table 1). Earlier meta-analyses were inconclusive or showed no association between FVL carriership and the risk of MI. A meta-analysis by Ye et al., including 12518 patients and 23374 controls, was the first to provide evidence that the presence of FVL is associated with a modest risk of MI (odds ratio [OR], 1.22; 95% CI, 1.10–1.35). These results were confirmed by a more recent meta-analysis, with a similar estimated risk of MI in FVL carriers (OR, 1.61; 95% CI, 1.30–1.98). Thus, based on the 2 latter studies, the concept of FVL representing a minor risk for MI in the general population can be concluded. Nevertheless, questions regarding the effect of FVL in important subpopulations (eg, in young individuals, smokers, women) remain to be answered. Especially young patients aged less than 45 years have been in the focus of the attention based on the assumption that if MI occurs at an earlier age, atherosclerosis may play a minor role in the pathomechanism and any risk related to an inherited prothrombotic state might be more easily detected.

One of the earliest case-control studies in this subgroup of interest was published by Rosendaal et al who analyzed 84 women with MI younger than 45 years of age and 388 control women. The authors observed that FVL was associated with a moderate risk of MI in young women (OR, 2.4; 95% CI, 1.0–5.9). A major finding of this study was that smoking carriers of FVL presented a 32-fold higher risk for MI as compared with nonsmoking noncarriers. The strong synergistic effect of smoking and thrombogenic risk factors (including FVL) was later confirmed in young men (<52 years of age) with MI as well, thus it is not sex specific. The moderately increased risk of MI in younger populations with FVL was not unequivocally concluded later by large case-control studies (n >500 cases; Table 2). However, a study including the largest number of young MI patients (n = 1880) did confirm a significant moderate risk. Studies on the risk of MI in young women carrying FVL are mostly contradictory, and no clear conclusion can be drawn based on these reports as yet.

**Prothrombin G20210A and the risk of myocardial infarction** Overview of prothrombin G20210A mutation

Prothrombin (FII), the precursor of thrombin, is a vitamin K–dependent glycoprotein. Thrombin is the central enzyme of coagulation. Besides its predominant role of transforming fibrinogen to fibrin, it enhances its own generation, it activates factor XIII that leads to the production of clots that are more resistant to fibrinolysis, but it also displays anticoagulant activity and attenuates its own generation via the thrombomodulin/PC pathway. During primary hemostasis, thrombin activates platelets via their proteinase-activated receptor 1 (PAR-1) or PAR-4, which provides a procoagulant phospholipid surface for coagulation to occur. Overall, 13 functions of thrombin have been described in hemostasis, and numerous nonhemostatic roles are also being discovered.
# TABLE 1  Summary of systematic reviews/meta‑analyses studying the association of factor V Leiden mutation with myocardial infarction

<table>
<thead>
<tr>
<th>Author</th>
<th>Studies, n</th>
<th>Cases/controls, n</th>
<th>Overall risk for MI</th>
<th>Age cutoff, y</th>
<th>Studies, n</th>
<th>Cases/controls, n</th>
<th>Young subgroup risk</th>
<th>Elderly subgroup risk</th>
<th>Risk in women</th>
<th>Risk in men</th>
<th>Conclusion: risk factor for MI?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dowaidar et al, 2010</td>
<td>41</td>
<td>7790/19276</td>
<td>OR, 1.61; 95% CI, 1.30–1.98; ( P &lt; 0.0001 ); ( I^2 = \text{ND} )</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Yes, moderate risk factor</td>
</tr>
<tr>
<td>Ye et al, 2006</td>
<td>53</td>
<td>12,518/23,374</td>
<td>OR, 1.22; 95% CI, 1.10–1.35; ( F = \text{ND} ); ( \rho_{\text{het}} = 0.01 )</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Yes, moderate risk factor</td>
</tr>
<tr>
<td>Middendorf et al, 2004</td>
<td>19</td>
<td>3655/5459</td>
<td>OR, 1.18; 95% CI, 0.97–1.43; ( F = \text{ND} ); ( \rho_{\text{het}} = 0.339 )</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>No significant effect</td>
</tr>
<tr>
<td>Kim et al, 2003</td>
<td>20</td>
<td>5313/14,047</td>
<td>OR, 1.10; 95% CI, 0.88–1.36; ( P = 0.41 ); ( F = \text{ND} ); ( \rho_{\text{het}} = \text{ND} )</td>
<td>&lt;55(^a)</td>
<td>14(^b)</td>
<td>624/1307</td>
<td>OR, 1.37; 95% CI, 1.06–1.97; ( P = 0.084 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = \text{ND} )</td>
<td>ND</td>
<td>OR, 1.79; 95% CI, 0.54–5.88; ( P = 0.34 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = \text{ND} )</td>
<td>OR, 1.22; 95% CI, 0.82–1.81; ( P = 0.33 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = \text{ND} )</td>
<td>No significant effect</td>
</tr>
<tr>
<td>Juul et al, 2002</td>
<td>19</td>
<td>4294/11,965</td>
<td>OR, 1.20; 95% CI, 1.03–1.39; ( P = 0.02 ); ( F = \text{ND} ); ( \rho_{\text{het}} = 0.61 )</td>
<td>&lt;50(^c)</td>
<td>7</td>
<td>764/8247</td>
<td>OR, 1.54; 95% CI, 1.07–2.22; ( P = 0.02 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = 0.69 )</td>
<td>ND</td>
<td>OR, 1.07; 95% CI, 0.56–2.04; ( P = 0.80 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = 0.07 )</td>
<td>OR, 1.15; 95% CI, 0.95–1.40; ( P = 0.15 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = 0.95 )</td>
<td>Potential, moderate risk factor in the overall population and in young individuals (&lt;55 y)</td>
</tr>
<tr>
<td>Boekholdt et al, 2001</td>
<td>12</td>
<td>2390/3547</td>
<td>OR, 1.29; 95% CI, 1.03–1.61; ( P = 0.02 ); ( F = \text{ND} ); ( \rho_{\text{het}} = \text{NS} )</td>
<td>&lt;55(^d)</td>
<td>6</td>
<td>1088/1454</td>
<td>OR, 1.34; 95% CI, 0.94–1.91; ( P = 0.10 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = \text{NS} )</td>
<td>ND</td>
<td>OR, 1.26; 95% CI, 0.94–1.67; ( P = 0.12 ); ( I^2 = \text{ND} ); ( \rho_{\text{het}} = \text{NS} )</td>
<td>ND</td>
<td>Potential, moderate risk factor in the overall population</td>
</tr>
</tbody>
</table>

OR (95% CI) was calculated based on an allele model, unless indicated otherwise.

\( a \) OR (95% CI) based on dominant model (FV Leiden carriers vs noncarriers); \( b \) Patients with arterial ischemic events (MI, ischemic stroke, or peripheral arterial occlusive disease)

Abbreviations: MI, myocardial infarction; ND, no data; NS, nonsignificant; \( \rho_{\text{het}} \), \( P \) value for heterogeneity; OR, odds ratio
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Embryonic development, wound healing, sepsis, cancer propagation, fibrosis, inflammation, etc (see Posma et al for a recent comprehensive review).

A common prothrombin gene mutation (FII 120210A), identified in 1996, is the second most common heritable thrombophilic defect. This gain-of-function mutation results in enhanced recognition of cleavage site, increased processing of the 3’ end, increased mRNA stability, mRNA accumulation and protein synthesis, without altering the structure of the protein. As a consequence, the presence of the mutation leads to moderately increased levels of prothrombin that can be readily converted to thrombin when necessary. Heterozygous individuals for FII 120210A have approximately 2- to 4-fold increased risk of VTE, while this risk in homozygotes is somewhat greater than 10-fold. Similarly to FVL, the prevalence of the FII 120210A mutation is dependent on ethnic background and shows geographic variability. FII 120210A, just as FVL, is almost absent in the natives of Africa, America, Australia, and east Asia, while in northern Europe, the allele frequency is approximately 1% to 2% and in Southern Europe it may reach up to 2% to 4%.

Studies on the role of prothrombin G20210A mutation in the development of myocardial infarction

The association of FII 20210A with the risk of MI has been studied in a number of conflicting and inconclusive reports. As the allele frequency of this inherited risk factor of thrombophilia is fairly low in the general population, only very large case-control studies or meta-analyses are expected to reliably assess its association with MI risk. Main results of published meta-analyses on the association of the FII G20210A polymorphism with MI are summarized in Table 3. Earlier meta-analyses including fewer than 5000 cases found no significant effect of the mutation in the general population, but pointed out the possibility that FII 20210A might be a potential risk factor for MI or arterial ischemic events in young individuals. A study by Ye et al including 8211 cases and 12356 controls, confirmed that the presence of FII 20210A represents a significant, but small risk for MI (OR, 1.25; 95% CI, 1.05–1.50). A more recent report by Li et al including a total of 33 studies with 13488 cases and 77085 controls showed a similar, significant association between FII 20210A and MI in the allele model (A vs G; OR, 1.43; 95% CI, 1.18–1.72) with almost identical results in the heterozygote (GA vs GG) or dominant model (GA+AA vs GG) as well. In this study, authors have performed subgroup analyses and found that the association remained significant in those of Caucasian origin. An important conclusion from this work is that the FII 20210A polymorphism is associated with MI risk in young patients (≤55 years), but not in

Table 2: Summary of large case-control studies (>500 cases) on the association of factor V Leiden mutation with the risk of myocardial infarction of the young

<table>
<thead>
<tr>
<th>Authors</th>
<th>Young cases/controls, n</th>
<th>Women/men in patient group, n</th>
<th>Young age cutoff, y</th>
<th>Risk in young individuals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tomaiuolo et al, 2012</td>
<td>955/909</td>
<td>362/593</td>
<td>&lt;45</td>
<td>Yes, moderate risk factor for MI in young women</td>
</tr>
<tr>
<td>Mannucci et al, 2010</td>
<td>1880/1880</td>
<td>210/1370</td>
<td>&lt;45</td>
<td>OR, 1.46; 95% CI, 0.99–2.22; p = 0.001</td>
</tr>
<tr>
<td>Atherosclerosis, Thrombosis and Vascular Biology Italian Study Group, 2003</td>
<td>1210/1210</td>
<td>149/1061</td>
<td>&lt;45</td>
<td>OR, 0.38; 95% CI, 0.15–0.92; p = 0.023</td>
</tr>
</tbody>
</table>

Abbreviations: see Table 1
<table>
<thead>
<tr>
<th>Authors</th>
<th>Studies, n</th>
<th>Cases/controls, n</th>
<th>Overall risk for MI</th>
<th>Age subgroups</th>
<th>Ethnicity subgroups</th>
<th>Gender subgroups</th>
<th>Conclusion: risk factor for MI?</th>
</tr>
</thead>
<tbody>
<tr>
<td>Li et al, 2017</td>
<td>33</td>
<td>13 488/77085</td>
<td></td>
<td>≤55</td>
<td>OR, 1.43; 95% CI, 1.18-1.72; P = 0.0002; F = 39.7; Phet &lt; 0.01</td>
<td></td>
<td>Yes, particularly in youngs (≤55 y) and in Caucasians</td>
</tr>
<tr>
<td>Ye et al, 2006</td>
<td>30</td>
<td>8 211/12 356</td>
<td></td>
<td>ND</td>
<td>ND</td>
<td>ND</td>
<td>Yes, moderate risk factor</td>
</tr>
<tr>
<td>Burzotta et al, 2004</td>
<td>13</td>
<td>3 687/6 078</td>
<td></td>
<td>≤55</td>
<td>OR, 1.19; 95% CI, 0.93-1.58; F = 23.1; Phet &lt; 0.01</td>
<td></td>
<td>Potential risk factor in young individuals (≤55 and ≤45 y)</td>
</tr>
<tr>
<td>Kim et al, 2003</td>
<td>14</td>
<td>4 887/7 840</td>
<td></td>
<td>&lt;55b</td>
<td>OR, 1.28; 95% CI, 0.94-1.73; F = ND; Phet = ND</td>
<td>ND</td>
<td>No, but risk factor for arterial ischemic events in young individuals (≤55 y)</td>
</tr>
<tr>
<td>Boekholdt et al, 2001</td>
<td>7</td>
<td>1 926/3 711</td>
<td></td>
<td>≤55</td>
<td>OR, 1.11; 95% CI, 0.79-1.56; P = 0.6; Fhet = NS</td>
<td></td>
<td>No, but borderline significant in young individuals (≤55 y)</td>
</tr>
</tbody>
</table>

OR (95% CI) was calculated based on an allele model (A vs G), unless indicated otherwise.

a OR (95% CI) based on dominant model (GA+AA vs GG); b Patients with arterial ischemic events (MI, ischemic stroke, or peripheral arterial occlusive disease); c Patients with unselected ischemic heart disease (MI, acute coronary syndrome, or unstable angina, or angiographically confirmed coronary artery disease).

Abbreviations: see TABLE 1.
it is plausible that MI events in young people are more likely to be attributable to genetic susceptibility to thrombotic risk than MI events in the elderly. It must be noted, however, that as compared with the number of younger individuals included in the analysis (n = 5294), only approximately half as many elderly patients with MI were available (n = 2174), demonstrating a possibility that elderly patients are lost through fatal MI or before the recruitment into case-control studies. A potential survival bias, in fact, is an important limitation of most case-control study designs published on this topic. In order to validate the above findings, large population (prospective) studies are warranted.

Another important question on the relation of common inherited thrombophilia risk factors (eg, FVL and FII20210A) and MI risk is whether the same risk applies to all forms of ATE (eg, MI, IS, peripheral arterial occlusive disease) in a patient, or is this effect substantially different between the various manifestations of ATE. In a systematic review by Maino et al\(^6\) most markers of hypercoagulability (including FVL) had a larger effect on the risk of IS as compared with MI, and this difference was more pronounced in young patients. Such predominant association with IS compared with MI was not as evident in elderly patients with MI were available (n = 2174), demonstrating a possibility that elderly patients are lost through fatal MI or before the recruitment into case-control studies. A potential survival bias, in fact, is an important limitation of most case-control study designs published on this topic. In order to validate the above findings, large population (prospective) studies are warranted.

Deficiencies of antithrombin, protein C, and protein S in arterial thrombotic disease Antithrombin deficiency Antithrombin is a serine protease inhibitor (serpin) and is the most important circulating inhibitor of blood coagulation.\(^5\) It primarily inactivates the thrombin-mediated formation of fibrin clot and the generation of thrombin by activated factor X. The mature form contains an N-terminal heparin-binding domain, a carbohydrate-rich domain, and a C-terminal serine protease-binding (reactive site) domain. According to its posttranslational modification, it has 2 isoforms differing from each other only in a glycosylation at Asn167.

Antithrombin deficiency was first described by Egberg\(^6\) in 1965, and the first functional AT defect, named AT Budapest, was reported by Sas et al\(^4\) in 1974. The deficiency is classified as type I (quantitative) and type II (qualitative) deficiency. The latter group includes deficiency of the heparin binding site (IIHS), the reactive site, and mutations leading to pleiotropic effects are also known.\(^5\) Antithrombin deficiency in general is associated with the highest risk of VTE, with the exception of the IIHBS type, which may confer a lower VTE risk in heterozygous state.\(^5\) Molecular genetic background of AT deficiency is highly heterogeneous, having more than 200 mutations listed in the HGMD database; however, there are some prevalent founder mutations, such as AT Cambridge II (p.Ala416Ser), AT Budapest 3 (ATBp3, p.Leu131Phe), and AT Basel (p.Pro73Leu).\(^51\)–\(^63\)

Deficiencies of proteins C and S Proteins C and S are vitamin K-dependent plasma glycoproteins.\(^6\) The domain structure of PC and PS is similar to other vitamin K-dependent coagulation factors, having a pre-pro leader sequence, a γ-glutamyl carboxylic acid domain, and epidermal growth factor–like domains. Protein C is a 2-chain protein in its mature form, and it contains an activation peptide domain and the serine protease domain. The latter is responsible for its inactivation effect to activated FV and FVIII. Protein S is a single-chain molecule that has a thrombin-sensitive region and a C-terminal region homologous to the sex hormone-binding globulin—like domain. The free form, approximately 40% of total PS, which is not bound to its natural binding protein (C4bBP) acts as a cofactor for APC. Protein S also has APC-independent anticoagulant effects that are not routinely tested in laboratories.\(^55\)–\(^56\) Both APC and PS have roles in several nonhemostasis processes.\(^6\) Protein S is involved in cell proliferation and survival, apoptosis, regulation of inflammatory cytokine release, atherosclerosis, vasculogenesis, and cancer development.\(^68\)

Deficiencies of PC and PS were first described by Griffin\(^69\) and Comp\(^70\) in the 1980s. Both deficiencies may be caused by mutations that lead to quantitative disease (type I) or qualitative alterations (type II). In PS deficiency, a laboratory phenotype of type III deficiency is also considered, where a normal total PS antigen level is associated with low free PS antigen and low APC cofactor activity. Several reports have proposed that type I and III deficiencies may be phenotypic variants of the same genetic disease.\(^71\) Molecular genetic background of deficiencies of PC and PS is heterogeneous with a much lower mutation detection rate than it is observed in AT deficiency, at least partly due to methodological issues.\(^64\) Founder mutations are also known in PC deficiency, and the PS Heerlen is a relatively frequent variant.\(^73\)–\(^74\)

Studies on deficiencies of antithrombin, protein C, and protein S in arterial thrombotic diseases As congenital deficiencies of AT, PC, and PS belong to rare diseases according to accepted definitions,\(^75\) well-designed, large clinical studies with an acceptable statistical power to find associations with MI risk are still lacking. Multicenter studies may overcome this problem; however, they are likely to be subjected to bias due to heterogeneous ethnicity. For this reason, here we focus not only on the association between deficiencies of AT, PC, and PS and the risk of MI, but the risk of other forms of arterial thrombotic events, mainly on IS as well. Nevertheless,
as of today, by searching the literature using the terms “antithrombin or protein C or protein S deficiency AND myocardial infarction or stroke or arterial thrombosis or arterial disease,” the majority of the published studies are single case reports.

Case reports on deficiencies of antithrombin, protein C, and protein S In rare diseases, well-written case reports may have an impact on the development of recommendations and clinical decision making if clinical and laboratory investigations provide evidence for the association between the questioned parameter and the disease. In this review case reports were considered based on the following selection criteria: objective confirmation of MI or IS diagnosis; data on AT, PC, or PS levels; publishing date is within the last 20 years. Publications on acquired deficiencies of AT, PC, or PS were excluded. A total of 34 case reports were consistent with the above criteria, the most important details of which are summarized in Supplementary material, Table S1.

Practically all patients in these case reports were young; all but one patient were younger than 50 years of age at the first presentation with ATE. This may suggest that hereditary deficiencies of the natural anticoagulants are severe enough to cause early onset of thrombotic complications; however, it may also reflect the present clinical practice of not investigating thrombophilia in elderly MI or IS patients. Male and female patients were equally affected with ATE according to these reports. The presence of well-known risk factors of ATE was excluded in most of the cases. Unfortunately, the timing of blood sampling for laboratory tests in some case reports has not been indicated, although it is recommended that at least 6 weeks should elapse after the acute event to exclude the influence of acute phase reaction. In most cases anticoagulant drugs were withdrawn before AT, PC, PS measurements. In studies without genetic diagnosis of deficiencies of AT, PC, or PS, the association between ATE and inherited form of the deficiency cannot be reliably confirmed. Family histories of the patients were not recorded in many cases, but there were some reports on multiple thrombotic episodes cosegregated with deficiencies of AT, PC, or PS, confirming the presence of hereditary deficiencies in those families. In conclusion, in young individuals with ATE, in the absence of classic risk factors, the chance of detecting underlying deficiencies of AT, PC, or PS was not negligible. In families with these deficiencies not only VTE but also ATE might develop, and combined thrombosis (ie, ATE plus VTE) might be common. It is to be noted, however, that in the case of IS, the presence of patent foramen ovale or arteriovenous fistula (which may reflect venous origin of thrombotic complications) was not unequivocally excluded in these case reports.

Case-control and cohort studies on deficiencies of antithrombin, protein C, and protein S The number of case-control studies on deficiencies of AT, PC, and PS is rather low. AT Cambridge II increased the risk of MI by 5.66-fold in a Spanish study,\textsuperscript{76} while it was without effect on IS in the Chinese population.\textsuperscript{77} On the contrary, the effect of rare thrombophilia was negligible in a study involving 255 MI cases and 400 control subjects in a recent study.\textsuperscript{78} When PC-deficient individuals (n = 34) and nonaffected individuals (n = 90) with ATE were compared, it was concluded that congenital PC deficiency contributed to an earlier onset of the disease.\textsuperscript{79}

There are 2 possible concepts when designing observational cohort studies in deficiencies of AT, PC, and PS. First, the detection of deficiencies (usually as part of thrombophilia screening) in ATE cohorts, and second, the detection of ATE in deficient cohorts. Both approaches have their rationale. It is to be considered, however, that the clinical research questions and the conclusions drawn from them are different.

In the first type of cohort studies, only a minor fraction of ATE patients are expected to have underlying deficiencies of AT, PC, or PS, particularly if the study involves a relatively low number of unselected patients. Therefore, these studies are often contradictory and inconclusive.\textsuperscript{80-83} Only a small proportion of ATE patients had deficiencies of AT, PC, or PS in most studies recruiting low number of individuals,\textsuperscript{84-85} except for young Brazilian individuals with IS (n = 130), where the prevalence of PS deficiency was 11.5%.\textsuperscript{86} In the most recent study including IS patients older than 65 years, 10 out of 89, 5 out of 99, and 8 out of 100 of tested patients had deficiencies of AT, PC, and PS, respectively. Nevertheless, this laboratory diagnosis led to a change in patient management in approximately one-third of the cases.\textsuperscript{87}

According to the second concept of study design, in which AT-, PC- or PS-deficient cohorts are investigated for ATE, the majority of published reports focus on AT deficiency. In this field, large cohort studies of 5 study groups are available.\textsuperscript{86,87,88-90} In the study by Alhenc-Gelas et al,\textsuperscript{88} 330 probands in France with AT deficiency and their relatives were recruited, and the prevalence of ATE was 4.8% in the whole population at a mean age of 38 years at the first presentation. The frequency of ATE was somewhat higher in type IIHBS (ATBp3, AT Basel, and AT Toyama), as compared with other types. In a Finnish study, where AT Basel was a frequent variant, nearly 12% of patients had experienced ATE and among them 5 patients had IS before the age of 45 years.\textsuperscript{89} Orlando registered ATE in 16 out of 82 individuals with type IIHBS AT deficiency, and AT Basel showed the strongest association with the disease as well.\textsuperscript{90} In a study from Germany, 72 type IIHBS patients were registered
out of 236 patients with AT deficiency and ATE was associated with type IIHBS (7 out of 72 individuals) more often than with other types (14 out of 236 individuals). In a Hungarian cohort of 246 AT-deficient individuals, ATE was associated more often with AT Basel (43%) and ATBp3 (10.5%) than with type I AT deficiency; the median age at first presentation was only 29 years in AT Basel and 33 years in AT Bp3 heterozygotes. In this cohort, ATE was reported in 3 children; 2 of them had IS and in one of them both MI and IS were found in the case history. In 2 smaller cohort studies involving 26 patients from southern Italy, only 4 patients were registered with type IIHBS AT deficiency and one of them had IS. In a study of 29 children from North America, no ATE was registered during the follow-up.

One can conclude from these studies that in patients with AT deficiency, ATE presents notably (approximately 20 years) earlier as compared with the general population. Another observation is that AT deficiency is a heterogeneous disease from the point of view of clinical manifestations, as the association with ATE seems to be the strongest in the case of type IIHBS.

As for PC deficiency, only 2 papers including a small number of patients were published. History of ATE was present in a considerable proportion of PC-deficient individuals in these studies. Moreover, PC deficiency significantly decreased the age at onset of symptoms of ATE in one of the studies. In hereditary PS deficiency, no association was found with ATE according to the single study available.

In a large family cohort study recruiting 552 individuals belonging to 84 different kindreds and having deficiencies of AT, PC, or PS, the hazard ratio for ATE was significantly higher in individuals with deficiency as compared with those who were nonaffected (HR, 2.3; 95% CI, 1.2–4.5). The risk of ATE was especially pronounced in young patients. When a subgroup analysis was performed, only deficiencies of PC and PS remained significant predictors of ATE under the age of 55 years.

In a cohort study including pooled data from 4 independent family cohort studies, the prevalence of ATE among 308 individuals with either AT, PC, or PS deficiency was 14%, which is high, although lower than that of VTE in the same group (56%). PC and PS deficiencies seemed to be more pronounced as compared with AT deficiency in ATE, and the risk conferred by deficiencies of AT, PC, and PS was only slightly higher than in the case of FVL and FII20210A. According to these results, inherited thrombophilia showed an association with ATE. Moreover, the study draws attention to a possible synergistic effect of traditional risk factors and thrombophilia in the pathogenesis of ATE.

Meta-analyses on deficiencies of antithrombin, protein C, and protein S Performing meta-analysis in any rare disease is a challenging task. This is at least in part caused by the difficulties of determining the predictors and outcome variables, which vary considerably throughout the individual studies. The heterogeneity in ethnicity is another issue, which makes the pooling of studies problematic. Literature search revealed only 2 meta-analyses on the association between deficiencies of AT, PC, or PS and the risk of ATE (IS). The first study concluded that thrombophilia is more common in children who had IS as compared with healthy ones. Protein C deficiency increased the risk of IS with an OR of 6.49 (95% CI, 2.95–14.27). In a more recent meta-analysis that investigated 1764 children with IS (including cerebral venous sinus thrombosis) and 2799 controls, a significant association was found between IS and each studied rare thrombophilia trait (AT deficiency, OR, 7.06; 95% CI, 2.44–22.42; PC deficiency, OR, 8.76; 95% CI, 4.53–16.96, and PS deficiency, OR, 3.20; 95% CI, 1.22–8.40).

In summary, based on the available case reports and results of population studies, laboratory investigation of rare inherited thrombophilia might be reasonable in selected patients with MI or IS. Factors that suggest underlying thrombosis risk factors are young age (<50 years), the lack of (or well-treated) risk factors of atherosclerosis, the lack of angiographic evidence of atherosclerosis (ie, the presence of a coronary or cerebral artery thrombus without major vessel stenosis), family member(s) with confirmed deficiencies of AT, PC, or PS, and family member(s) who had VTE or ATE at a young age.

**Conclusion** In conclusion, the risk of MI in the case of a patient with common inherited thrombophilia (FVL and FII20210A) is low or moderate at most. In patients with deficiencies of AT, PC, or PS, the risk of MI or other ATEs is not negligible, and seem to occur at a younger age as compared with the general population. In general, a personalized clinical approach is suggested when testing for inherited thrombophilia in a patient with MI. If the diagnosis of inherited thrombophilia influences the clinical management of the patient and helps avoid the recurrence of thrombosis, it is definitely worth testing. Whether or not the patient would benefit from antithrombotic prophylaxis must be decided on the total thrombotic risk. The concept of combined antiplatelet and anticoagulant agents for the secondary prevention of atherothrombotic events have been revisited in the era of direct oral anticoagulants as they have lower risk of intracerebral bleeding as compared with vitamin K antagonists. Nevertheless, future studies and guidance are warranted to help identify patients who will benefit most from thrombophilia testing and altered secondary prevention.
SUPPLEMENTARY MATERIAL
Supplementary material is available at www.mp.pl/kardiologiapolksa.

ARTICLE INFORMATION
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CONFLICT OF INTEREST
None declared.

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REFERENCES


