Role of inherited thrombophilic profile on survival of patients with sepsis

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ABSTRACT

The existence of various coagulation and/or fibrinolytic system disorders (such as inherited thrombophilia) in patients with sepsis could possibly modify host response to infection as well as patient outcome. The aim of the study is to investigate inherited thrombophilic profile in patients with sepsis. Eighty-three patients with sepsis admitted at the Department of Internal Medicine of the University General Hospital of Patras, Greece were included. Thrombophilic profile (factor V G1691A (Leiden), factor V H1299R (R2), prothrombin G20210A, MTHFR C677T, MTHFR A1298C, factor XIII V34L, β-fibrinogen-455 G-A and plasminogen activator inhibitor (PAI)-1 4G/5G) was evaluated using the cardiovascular diseases (CVD) StripAssay based on DNA isolation, PCR and reverse hybridisation. Data were collected from patients' chart reviews. Seventy patients (84.3%) of the 83 enrolled had at least one thrombophilic mutation. The most common mutations were heterozygous for β-fibrinogen-455 G-A (43.4%), heterozygous for factor XIII V34L (32.5%), PAI-1 4G/4G (26.5%), homozygous MTHFR C677T (22.9%), heterozygous factor V H1299R (R2) (13.3%) and homozygous MTHFR A1298C (12.0%). A 30-day mortality was 14.5%. Multivariate analysis revealed that mortality was independently associated with Simplified Acute Physiology Score II score on admission, pneumonia and fibrinogen on admission. Nine patients (10.8%) developed septic shock. Coagulation disorders on admission, bacteraemia and PAI-1 genotype 5G/5G were independently associated with development of septic shock. The presence of thrombophilic mutations in patients with sepsis may affect their clinical response, and future studies are needed in order to elucidate the role of isolated thrombophilic mutations in patients with sepsis or septic shock.

Significance of this study

What is already known about this subject?
- Coagulation disorders are important causes of sepsis morbidity and mortality.
- Haemostatic gene polymorphisms' role on sepsis morbidity have been studied with conflicting results.
- 4G/4G genotype of plasminogen activator inhibitor 1 was associated with increased risk for complications during meningococcal disease.

What are the new findings?
- Low fibrinogen levels were independently associated with mortality.
- Mortality was not influenced by thrombophilic profile.
- 5G/5G genotype of the plasminogen activator inhibitor 1 was independently associated with development of septic shock.

How might these results change the focus of research or clinical practice?
- Studies with large number of participants are needed in order to clarify the impact of haemostatic gene polymorphisms and especially plasminogen activator inhibitor 1 on septic shock development and mortality among patients with sepsis.

INTRODUCTION

Sepsis remains a major public health issue, which is characterised by a dysregulated host response to infection and leads to increased morbidity and mortality.[1 2] It is well known that sepsis is a life-threatening organ dysfunction caused by a dysregulated host response to infection.[1] Particularly in sepsis, the coagulation cascade is activated, while the anticoagulant mechanisms and fibrinolysis are downregulated.[3–6] In turn, the dysregulation of coagulation system and subsequent thrombin deposition represent a crucial part of the host response to infection that considerably affects severity and clinical course of sepsis.[3 6–9] The clinical expression of such dysregulation is deep vein thrombosis and pulmonary embolism[10]; therefore, it is strongly recommended to use unfractionated heparin or low-molecular-weight heparin in order to prevent aforementioned complications.[11]

The role of haemostatic gene polymorphisms in sepsis-induced coagulopathy disorders and subsequently to mortality has been studied especially in animal models, with sometimes conflicting results.[8 12] The most commonly recognised mutation in haemostatic gene is the factor V Leiden (fVL) mutation and it is thoroughly studied in meningococcal disease. In a previous study, patients with heterozygous fVL

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Table 1 Univariate analysis of predictors of 30-day mortality of patients admitted to the Department of Internal Medicine with sepsis

Characteristics	Survivors (71 patients)	Non survivors (12 patients)	P value
Demographics			
Gender (male)	38 (53.5%)	3 (25.0%)	0.116
Age (years)	65.4±20.2	78.5±14.4	0.011
Comorbidities			
Charlson Comorbidity Index	3.0±2.0	5.3±2.6	0.003
COPD	8 (11.3%)	3 (25.0%)	0.194
Cerebrovascular accident	8 (11.3%)	2 (16.7%)	0.633
Diabetes mellitus	15 (21.1%)	0 (0.0%)	0.112
Malignancy (haematological or solid organ)	5 (7.0%)	2 (16.7%)	0.266
Admission data			
Duration of symptoms before admission (days)	2.9±3.0	1.8±1.8	0.100
Leucocytes (10^9/L)	16.5±5.9	12.3±4.4	0.025
C reactive protein (mg/dL) on admission	14.3±8.6	16.2±8.7	0.472
Temperature (°C)	38.7±0.6	37.8±1.5	0.035
SAPS II Score on admission	36.9±13.4	61.1±15.0	<0.001
SOFA Score on admission	4.3±2.2	7.1±1.9	<0.001
Acute kidney injury	11 (15.5%)	7 (58.3%)	0.003
LOS (days)	11.0±9.1	12.6±14.2	0.984
Infection severity			
Sepsis	66 (93.0%)	8 (66.7%)	
Septic shock	5 (7.0%)	4 (25.0%)	0.022*
Type of infection			
Pneumonia	19 (26.8%)	7 (58.3%)	0.043†
Urinary tract infection	32 (45.1%)	4 (33.3%)	
Abdominal infection	13 (18.3%)	0 (0.0%)	
Other‡	7 (9.8%)	1 (8.3%)	
Bacteraemia	12 (16.9%)	3 (25.0%)	0.447
Microbiological confirmed	28 (39.4%)	6 (50.0%)	1.000
Gram-positive	4 (5.6%)	1 (8.3%)	
Gram-negative	24 (33.8%)	5 (41.7%)	
Coagulation data			
Coagulation disorders on admission	7 (9.9%)	3 (25.0%)	0.154
INR	1.2±0.2	1.3±0.2	0.027
PTT (s)	34.6±7.9	37.7±10.5	0.375
Platelets (10^9/L)	246.1±117.2	228.5±75.9	0.746
Fibrinogen (g/L)	650.6±176.9	518.1±210.1	0.079
D-dimers (ng/mL)	2757.6±2664.5	3731.0±3139.3	0.392
Coagulation disorders during 3rd day of hospitalisation	4 (5.6%)	4 (33.3%)	0.013
INR	1.1±0.1	1.3±0.2	0.001
PTT (s)	37.2±6.3	37.8±8.8	0.736
Platelets (10^9/L)	235.8±106.4	174.8±74.7	0.078
Fibrinogen (g/L)	668.7±169.8	533.5±183.3	0.042
D-dimers (ng/mL)	2564.3±1954.1	5748.0±3593.9	0.012
Coagulation disorders during seventh day of hospitalisation§	2 (2.8%)	3 (25.0%)	0.020
INR	1.1±0.1	1.3±0.3	0.012
PTT (s)	34.7±5.7	40.6±13.7	0.349
Platelets (10^9/L)	314.5±126.0	173.1±52.1	0.001
Fibrinogen (g/L)	607.4±170.4	432.9±202.2	0.017
D-dimers (ng/mL)	2927.5±1992.1	5814.4±3772.6	0.049

*Comparison between patients with septic shock against those with sepsis.
†Comparison between patients with pneumonia against all other causes.
‡Skin and soft-tissue infections and endocarditis.
§Comparison among 56 patients.
COPD, chronic obstructive pulmonary disease; INR, international normalized ratio; LOS, length of stay; PTT, partial thromboplatin time; SAPS II, Simplified Acute Physiology Score II; SOFA, sequential organ failure assessment.

mutation had increased risk for complications of purpura fulminans during meningococcal disease, while no association with mortality was found.[13] Another gene polymorphism associated with increased risk for complications of purpura fulminans during meningococcal disease and also mortality is the 4G/4G genotype of the plasminogen activator inhibitor (PAI)-1.[14]

Existence of inherited thrombophilia in patients with sepsis could possibly affect coagulation system, host response to infection and subsequently clinical course of sepsis. Data on the role of haemostatic gene polymorphisms among patients with sepsis are scarce. The aim of the present study is to investigate the role of such polymorphisms on mortality and development of septic shock.

MATERIALS AND METHODS

Study population

The present study is a prospective one and was performed at the Department of Internal Medicine of the University General Hospital, Patras during a 12-month period. Inclusion criteria were age ≥18 years and admission at the Department of Internal Medicine with sepsis. Exclusion criteria were hospitalisation for less than 24 hours, HIV infection, neutropaenia, moderate to severe cirrhosis, as well as patients receiving immunosuppressive agents or anticoagulant therapy.

Patients were enrolled in the study, within 24 hours of admission. Primary outcome was a 30-day mortality. A secondary analysis was performed in order to assess the role of thrombophilic mutation in the development of septic shock. Data (epidemiological data, comorbidities, severity on admission, laboratory and radiological results) were obtained by patients' chart reviews. Type of infection was defined according to Centers for Disease Control and Prevention (CDC) definition.[2] According to the Society of Critical Care Medicine (SCCM)/ European Society of Intensive Care Medicine (ESICM)/ American College of Chest Physicians (ACCP)/ American Thoracic Society (ATS)/ Surgical Infection Society (SIS) consensus conference, sepsis was defined as infection with systemic inflammatory response syndrome, and septic shock as sepsis with arterial hypotension despite adequate fluid resuscitation.[15]

Genotypic analysis

Blood samples in ethylenediaminetetraacetic acid (EDTA)-containing tubes for DNA studies were collected on

Table 2 Thrombophilic profile of patients admitted to the department of internal medicine with sepsis according to mortality or severity of infection

Characteristics	Survivors (71 patients)	Non survivors (12 patients)	P value	Sepsis (74 patients)	Septic shock (9 patients)	P value
FV Leiden						
Heterozygote	4 (5.6%)	0 (0.0%)		4 (5.4%)	0 (0.0%)	
Homozygote	0 (0.0%)	0 (0.0%)	1.000*	0 (0.0%)	0 (0.0%)	1.000*
FV H1299R (R2)						
Heterozygote	9 (12.7%)	2 (16.7%)		8 (10.8%)	3 (33.3%)	
Homozygote	1 (1.4%)	2 (16.7%)	0.053*	3 (4.1%)	0 (0.0%)	1.000*
FII-prothrombin (G20210A)						
Heterozygote	3 (4.2%)	0 (0.0%)		3 (4.1%)	0 (0.0%)	
Homozygote	0 (0.0%)	0 (0.0%)	1.000*	0 (0.0%)	0 (0.0%)	1.000*
FXIII (V34L)						
Heterozygote	25 (35.2%)	2 (16.7%)		25 (33.8%)	2 (22.2%)	
Homozygote	1 (1.4%)	0 (0.0%)	1.000*	1 (1.4%)	0 (0.0%)	1.000*
β-fibrinogen						
Heterozygote	29 (40.8%)	7 (58.3%)		32 (43.2%)	4 (44.4%)	
Homozygote	6 (8.5%)	0 (0.0%)	0.356*	5 (6.8%)	1 (11.1%)	0.509*
MTHFR (C677T)						
Heterozygote	28 (39.4%)	4 (33.3%)		28 (37.8%)	4 (44.4%)	
Homozygote	14 (19.7%)	5 (41.7%)	0.134*	18 (24.3%)	1 (11.1%)	0.677*
MTHFR (A1298C)						
Heterozygote	28 (39.4%)	4 (33.3%)		27 (36.5%)	5 (55.6%)	
Homozygote	10 (14.1%)	0 (0.0%)	0.344*	9 (12.2%)	1 (11.1%)	1.000*
PAI-1 genotype						
4G/4G	18 (25.4%)	4 (33.3%)	0.724†	22 (29.7%)	0 (0.0%)	0.104†
4G/5G	31 (43.7%)	3 (25.0%)	0.343†	32 (43.2%)	2 (22.2%)	0.297†
5G/5G	22 (31.0%)	5 (41.7%)	0.514†	20 (27.0%)	7 (77.8%)	0.005†
No of mutations‡	1.7±1.1	1.8±1.2	0.679	1.7±1.1	1.5±1.2	0.571

*Comparison among homozygote against heterozygote or no mutation.
†Comparison among each genotype against both other genotypes.
‡Mutation defined as FV G1691A (Leiden) homozygous or heterozygous, FV H1299R (R2) homozygous or heterozygous, FII-prothrombin (G20210A) homozygous or heterozygous, FXIII V34L homozygous or heterozygous, β-fibrinogen homozygous or heterozygous, PAI-1genotype 4G/4G, MTHFR C677T homozygous, MTHFR A1298C homozygous; presence of homozygous mutation was calculated as one mutation, as was heterozygous.
FV, factor V; PAI-1, plasminogen activator inhibitor-1.

enrolment. The following thrombophilic mutations were evaluated using the cardiovascular diseases (CVD) StripAssay T 4–360 (ViennaLab Labordiagnostika): factor V G1691A, factor V H1299R (R2), prothrombin G20210A, MTHFR C677T, MTHFR A1298C, factor XIII V34L, β-fibrinogen-455 G-A and PAI-1 4G/4G. The procedure was based on DNA isolation, PCR and reverse hybridization.

Statistical analysis

Statistical analysis was performed using SPSS V.23.0 (SPSS). The Fisher's exact test or X^2 test was employed for categorical variables, while, Mann-Whitney U test or t-test for continuous ones, as appropriate. Multiple logistic regression analysis was used to calculate unadjusted ORs and 95% CIs. Factors contributing to multicollinearity were excluded from the multivariate analysis. Statistical significance was established at $p<0.05$.

RESULTS

Overall 83 patients were enrolled in our study. The most common types of infections were urinary tract infection (36 patients, 43.4%), pneumonia (26 patients, 31.3%) and abdominal infections (13 patients, 15.7%), while the remaining eight infections were skin and soft-tissue infections and endocarditis. Fifteen patients (18.1%) developed bacteraemia. Only 34 infections (41.0%) had microbiological confirmation of the pathogen; 29 were caused by gram-negative bacteria and 5 were gram-positive.

Seventy patients (84.3%) had at least one thrombophilic mutation. Three patients (3.6%) had four mutations, 19 (22.9%) had three mutations and 25 (30.1%) had two. The most common mutations were heterozygous for β-fibrinogen-455 G-A (36; 43.4%), heterozygous for factor XIII V34L (27; 32.5%), PAI-1 4G/4G (22; 26.5%), homozygous MTHFR C677T (19; 22.9%), heterozygous factor V H1299R (R2) (11; 13.3%) and homozygous MTHFR A1298C (10; 12.0%).

A 30-day mortality was 14.5% (12 patients). Univariate analysis of risk factors for mortality is depicted in tables 1 and 2. No significant differences were found in a 30-day mortality among patients with and without thrombophilic mutations. Multivariate analysis revealed that Simplified Acute Physiology Score II on admission (p=0.014; OR 1.2, 95% CI 1.0 to 1.4), pneumonia (p=0.016; OR 33.5, 95% CI

Table 3 Univariate analysis of differences among patients with sepsis or septic shock admitted to the department of internal medicine

Characteristics	Sepsis (74 patients)	Septic shock (9 patients)	P value
Demographics			
Gender (male)	38 (51.4%)	3 (33.3%)	0.483
Age (years)	65.7±20.0	79.8±15.4	0.016
Comorbidities			
Charlson Comorbidity Index	3.1±2.1	5.0±2.4	0.016
COPD	10 (13.5%)	1 (11.1%)	1.000
Cerebrovascular accident	8 (10.8%)	2 (22.2%)	0.296
Diabetes mellitus	13 (17.6%)	2 (22.2%)	0.663
Malignancy (haematological or solid organ)	6 (8.1%)	1 (11.1%)	0.567
Admission data			
Duration of symptoms before admission (days)	2.8±2.9	2.0±2.1	0.224
Leucocytes (10^9/L)	16.3±5.7	12.7±6.5	0.128
C reactive protein (mg/dL) on admission	14.9±8.7	12.9±8.4	0.662
Temperature (°C)	38.7±0.7	37.9±1.3	0.093
SAPS II Score on admission	37.9±14.5	61.1±13.5	<0.001*
SOFA Score on admission	4.0±1.9	8.4±2.2	<0.001*
Acute kidney injury	11 (14.9%)	8 (88.9%)	<0.001
LOS (days)	10.9±9.0	13.2±16.4	0.866
Type of infection			
Pneumonia	23 (31.1%)	3 (33.3%)	1.000†
Urinary tract infection	31 (41.9%)	5 (55.6%)	
Abdominal infection	13 (17.6%)	0 (0.0%)	
Other‡	7 (9.5%)	1 (11.1%)	
Bacteraemia	11 (14.9%)	4 (44.4%)	0.052
Microbiological confirmed	30 (40.5%)	4 (44.4%)	1.000
Gram-positive	3 (4.1%)	2 (22.2%)	
Gram-negative	27 (36.5%)	2 (22.2%)	
Coagulation disorders on admission	6 (8.1%)	4 (44.4%)	0.010
INR	1.2±0.2	1.3±0.3	0.115
PTT (s)	34.4±7.9	40.3±10.0	0.079
Platelets (10^9/L)	250.0±110.6	191.0±116.2	0.110
Fibrinogen (g/L)	650.1±183.7	489.5±139.5	0.297
D-dimers (ng/mL)	2781.6±2684.1	3767.5±3131.7	0.392

*Factors were not included in the multivariate analysis.
†Comparison between patients with pneumonia against all other causes.
‡Skin and soft-tissue infections and endocarditis.
SAPS II, Simplified Acute Physiology Score II.

1.9 to 580.6) and fibrinogen (p=0.032; OR 0.988, 95% CI 0.977 to 0.999) were associated with a 30-day mortality.

Nine patients (10.8%) developed septic shock necessitating admission at the intensive care unit. Univariate analysis of risk factors for development of septic shock is depicted in tables 2 and 3. Coagulation disorders on admission (p=0.015; OR 10.4, 95% CI 1.6 to 68.4), bacteraemia (p=0.026; OR 15.3, 95% CI 1.7 to 136.1) and PAI-1 genotype 5G/5G (p=0.019; OR 15.1, 95% CI 1.6 to 145.4) were independently associated with the development of septic shock.

DISCUSSION

The majority of patients with sepsis have relevant coagulation abnormalities, ranging from mild decrease in platelet count or weak prolongation of clotting times, to more severe coagulation disorders and sometimes disseminated intravascular coagulation.[6 16 17] These coagulation abnormalities could be explained by an extensive bidirectional interaction between sepsis and coagulation.[6 9 18 19] Moreover, proinflammatory cytokines and chemokines are the main mediators in this interaction between sepsis and coagulation.[18]

As previously shown, pneumonia as compared with other types of infections was associated with higher mortality.[20–22] Even though bacteraemia was not associated with worst outcome, it was more common among patients that developed septic shock. Gram-negative pathogens predominated in our cohort, since urinary tract infections were the most common type of infection for which microbiological documentation is higher as comparison to other types of infections and especially pneumonia.

In the present study, a high percentage of patients (84.3%) carried at least one thrombophilic mutations, which is in accordance with previous studies conducted in general population, as well as in patients with sepsis.[23–28] Moreover, percentages of individual mutations are comparable to that reported from studies deriving from Europe. Even though most of patients included carried thrombophilic mutations, mortality was influenced by neither individual mutations nor accumulation of such mutations.

Fibrinogen's role in the survival from infection and especially sepsis has been previously established, with higher fibrinogen levels being associated with higher survival.[29] An association between higher plasma fibrinogen levels and −455 G/A polymorphism in the promoter region of the fibrinogen-beta gene has been shown by previous studies.[30] Even though we found that lower levels of fibrinogen led to higher mortality, the presence of heterozygous (n=34 patients) or homozygous (n=6)–455 G/A polymorphism was not associated with better outcome.

PAI-1 is a crucial inhibitor of fibrinolysis by inhibiting plasminogen activator, so high circulating levels lead to disseminated intravascular coagulation and organ dysfunction and are associated with worst outcome in infected patients and development of septic shock.[31] The most common studied polymorphism was a deletion/insertion (4G/5G) one in the promoter region of aforementioned gene. The 4G allele (four guanine bases) has been consistently associated with higher levels of PAI-1, thus leading to higher mortality among septic patients.[32 33] To the best of our knowledge, this is the first study to report that 5G/5G genotype, as compared with both 4G/4G and 4G/5G, was associated with a higher risk for septic shock development, even though no association with mortality was found. A possible explanation for our results may be that production of PAI-1 is influenced apart from genetic factors by many other determinants including metabolic factors, lifestyle, renin–angiotensin system and other inflammatory mediators.[32 34]

From the remaining studied mutations (factor V G1691A (Leiden), factor V H1299R (R2), prothrombin G20210A, MTHFR C677T, MTHFR A1298C and factor XIII V34L),

fVL mutation was the most thoroughly investigated in experimental murine models and clinical sepsis studies with controversial results. In a Danish population-based study, fVL mutation was associated with an increased risk of a 28-day mortality among septic patients,[35] while Kerlin *et al*[28] showed a survival advantage among heterozygous fVL septic patients as compared with non-carriers. Nevertheless, a recent meta-analysis showed no association between fVL mutation and mortality risk.[12] In the present study, fVL mutation had no influence on mortality, but only four patients (5.6%) were heterozygotes.

The study had some limitations. The development of sepsis and organ dysfunction after an infection has to be considered as an individualised process. The sample size in our study was relatively small for a condition that varies substantially in its clinical spectrum and severity. Even though most of the patients included had at least one thrombophilic mutation, the effect of some rare polymorphisms could not be properly evaluated. Second, no measurement of PAI-1 circulating levels was conducted in order to evaluate if the effect of 5G/5G genotype on septic shock development was a random association or was associated with higher circulating PAI-1 levels.

Although coagulopathy disorders, such as low fibrinogen levels, play an important role in sepsis-related mortality, the role of individual or combination of thrombophilic mutations remains unclear. PAI-1 5G/5G genotype, as compared with both 4G/4G and 4G/5G, was associated with higher risk for septic shock development, while none of the studied polymorphisms influenced mortality. Consequently, future studies with large number of participants are needed in order to clarify the impact of haemostatic gene polymorphisms on mortality among the patients with sepsis or septic shock.

Contributors MM and MK conceived the idea and experimental design. AG and MP-O collected the patients' data. AG collected the blood samples. AG and MK conducted the molecular experiments. MM supervised the project. MP-O and AG performed the analysis and interpreted the results. AG and MP-O wrote the manuscript. All authors contributed to manuscript revision, read and approved the submitted version.

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REFERENCES

1 Singer M, Deutschman CS, Seymour CW, *et al*. The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). *JAMA* 2016;315:801–10.
2 Horan TC, Andrus M, Dudeck MA. CDC/NHSN surveillance definition of health care-associated infection and criteria for specific types of infections in the acute care setting. *Am J Infect Control* 2008;36:309–32.
3 Schouten M, Wiersinga WJ, Levi M, *et al*. Inflammation, endothelium, and coagulation in sepsis. *J Leukoc Biol* 2008;83:536–45.
4 Munford R, Suffredini A. Sepsis, severe sepsis, and septic Shock. In: Mandell G, Bennett J, Dolin R, eds. *Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases*: Elsevier, 2010:987–1010.
5 Semeraro N, Ammollo CT, Semeraro F, *et al*. Sepsis, thrombosis and organ dysfunction. *Thromb Res* 2012;129:290–5.
6 Levi M, Schultz M, van der Poll T. Sepsis and thrombosis. *Semin Thromb Hemost* 2013;39:559–66.
7 Aird WC. Vascular bed-specific hemostasis: role of endothelium in sepsis pathogenesis. *Crit Care Med* 2001;29:S28–34. discussion S34-5.
8 Texereau J, Pene F, Chiche JD, *et al*. Importance of hemostatic gene polymorphisms for susceptibility to and outcome of severe sepsis. *Crit Care Med* 2004;32:S313–9.
9 Levi M, van der Poll T, Büller HR. Bidirectional relation between inflammation and coagulation. *Circulation* 2004;109:2698–704.
10 Kaplan D, Casper TC, Elliott CG, *et al*. VTE Incidence and Risk Factors in Patients With Severe Sepsis and Septic Shock. *Chest* 2015;148:1224–30.
11 Rhodes A, Evans LE, Alhazzani W, *et al*. Surviving Sepsis Campaign: International Guidelines for Management of Sepsis and Septic Shock: 2016. *Intensive Care Med* 2017;43:304–77.
12 Zhang J, He Y, Song W, *et al*. Lack of association between factor V Leiden and sepsis: a meta-analysis. *Clin Appl Thromb Hemost* 2015;21:204–10.
13 Kondaveeti S, Hibberd ML, Booy R, *et al*. Effect of the Factor V Leiden mutation on the severity of meningococcal disease. *Pediatr Infect Dis J* 1999;18:893–6.
14 Binder A, Endler G, Müller M, *et al*. 4G4G genotype of the plasminogen activator inhibitor-1 promoter polymorphism associates with disseminated intravascular coagulation in children with systemic meningococcemia. *J Thromb Haemost* 2007;5:2049–54.
15 Levy MM, Fink MP, Marshall JC, *et al*. SCCM/ESICM/ACCP/ATS/SIS International Sepsis Definitions Conference. *Crit Care Med* 2001;2003:1250–6.
16 Levi M. The coagulant response in sepsis. *Clin Chest Med* 2008;29:627–42.
17 Levi M, van der Poll T, Schultz M. New insights into pathways that determine the link between infection and thrombosis. *Neth J Med* 2012;70:114–20.
18 Levi M, van der Poll T. Two-way interactions between inflammation and coagulation. *Trends Cardiovasc Med* 2005;15:254–9.
19 Levi M, van der Poll T. Coagulation and sepsis. *Thromb Res* 2017;149:38–44.
20 Falagas ME, Korbila IP, Kapaskelis A, *et al*. Trends of mortality due to septicemia in Greece: an 8-year analysis. *PLoS One* 2013;8:e67621.
21 Papadimitriou-Olivgeris M, Aretha D, Zotou A, *et al*. The Role of Obesity in Sepsis Outcome among Critically Ill Patients: A Retrospective Cohort Analysis. *Biomed Res Int* 2016;2016:1–9.
22 Mansur A, Klee Y, Popov AF, *et al*. Primary bacteraemia is associated with a higher mortality risk compared with pulmonary and intra-abdominal infections in patients with sepsis: a prospective observational cohort study. *BMJ Open* 2015;5:e006616.
23 Karakantza M, Androutsopoulos G, Mougiou A, *et al*. Inheritance and perinatal consequences of inherited thrombophilia in Greece. *Int J Gynaecol Obstet* 2008;100:124–9.
24 Franco RF, Reitsma PH. Genetic risk factors of venous thrombosis. *Hum Genet* 2001;109:369–84.
25 Buchanan GS, Rodgers GM, Ware Branch D. The inherited thrombophilias: genetics, epidemiology, and laboratory evaluation. *Best Pract Res Clin Obstet Gynaecol* 2003;17:397–411.
26 Middeldorp S, Levi M. Thrombophilia: an update. *Semin Thromb Hemost* 2007;33:563–72.
27 Tsantes AE, Tsangaris I, Bonovas S, *et al*. The effect of four hemostatic gene polymorphisms on the outcome of septic critically ill patients. *Blood Coagul Fibrinolysis* 2010;21:175–81.
28 Kerlin BA, Yan SB, Isermann BH, *et al*. Survival advantage associated with heterozygous factor V Leiden mutation in patients with severe sepsis and in mouse endotoxemia. *Blood* 2003;102:3085–92.
29 Kaspereit F, Doerr B, Dickneite G. The effect of fibrinogen concentrate administration on coagulation abnormalities in a rat sepsis model. *Blood Coagul Fibrinolysis* 2004;15:39–43.
30 Manocha S, Russell JA, Sutherland AM, *et al*. Fibrinogen-beta gene haplotype is associated with mortality in sepsis. *J Infect* 2007;54:572–7.
31 Tipoe TL, Wu WKK, Chung L, *et al*. Plasminogen Activator Inhibitor 1 for Predicting Sepsis Severity and Mortality Outcomes: A Systematic Review and Meta-Analysis. *Front Immunol* 2018;9:1218.
32 Hermans PW, Hazelzet JA. Plasminogen activator inhibitor type 1 gene polymorphism and sepsis. *Clin Infect Dis* 2005;41(Suppl 7):S453–8.
33 Li L, Nie W, Zhou H, *et al*. Association between plasminogen activator inhibitor-1 -675 4G/5G polymorphism and sepsis: a meta-analysis. *PLoS One* 2013;8:e54883.
34 Horrevoets AJ. Plasminogen activator inhibitor 1 (PAI-1): in vitro activities and clinical relevance. *Br J Haematol* 2004;125:12–23.
35 Benfield TL, Dahl M, Nordestgaard BG, *et al*. Influence of the factor V Leiden mutation on infectious disease susceptibility and outcome: a population-based study. *J Infect Dis* 2005;192:1851–7.