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Research article

PREDICTING RISKS OF PROTHROMBOTIC READINESS UNDER COVID-19 USING GENETIC TESTING

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COVID-19 poses a significant hazard as regards decompensation of underlying chronic diseases, specific damage to the cardiovascular system, and a high risk of negative health outcomes such as thrombotic events. The coronavirus infection pathogenesis is rather complicated and has not been studied yet; this is largely due to peculiar features of the virus and the initial state of homeostasis in a patient.

In this study, our aim was to analyze molecular-genetic markers of homeostasis in patients with the new coronavirus infection COVID-19 as a prognostic trigger of developing pro-thrombotic readiness.

Hospitalized patients with COVID-19 were chosen as study objects. We performed molecular-genetic analysis of basic genes significant for homeostasis including several factors such as V (rs6025), II (rs1799963), I (rs1800790), VII (rs6046), XIII A1 (rs5985)), IGN A2 (rs1126643), IGN B3 (rs5918), and PAI-1 (rs1799889). The thrombinemia severity was identified by thrombin generation tests using the Ceveron®alpha automated coagulation analyzer with TGA-module.

Allelic variants of PAI-1, prothrombin (FII), and fibrinogen (FI) determined high thrombinemia as per the thrombin kinetics test (endogenous thrombin potential (AUC), peak thrombin concentration (peak-thrombin), time necessary to reach thrombin peak (tPeak), levels of fibrinogen and D-dimer) in COVID-19 patients during the entire hospitalization. We established that elevated thrombin generation becoming apparent through elevated levels of endogenous thrombin potential (AUC) might be a prognostic indicator of the pro-thrombotic state in patients with genetic polymorphisms of PAI-I and fibrinogen.

The study results indicate that pro-thrombotic readiness is determined genetically in case COVID-19 patients have allelic variants in PAI-I, prothrombin (factor II) and fibrinogen (factor I) genes.

Keywords: *COVID-19, genotype, risk, mutation, thrombinemia, polymorphism, thrombin, thrombosis.*

The COVID-19 pandemic created a wide-scale crisis in public healthcare causing millions of deaths all over the world. Clinical symptoms of the infection vary between mild and critical; pneumonia with acute respiratory distress syndrome and organ dysfunction is the most frequent severe case of the disease. A new outbreak of the coronavirus infection, in case the disease is severe, poses serious threats as regards decompensation of some initial chronic diseases, specific lesion of the cardiovascular system, a high risk of unfavorable outcomes such as thrombotic events [1]. Both foreign and Russian studies report high frequency of thrombotic events in hospitalized patients with non-critical COVID-19 despite conventional thrombotic prophylaxis. The coronavirus infection has rather complicated pathogenesis that has not yet been studied; it largely depends on peculiarities of the virus itself and the initial state of a patient's homeostasis [2]. Anti-inflammatory cytokines are known to be able to stimulate expression of tissue thromboplastin on immune cells and activate coagulation under COVID-19. Inflammation-induced endothelial dysfunction further accelerates prothrombotic readiness and elevated thrombin generation, inhibits fibrinolysis activity by reducing activity of urokinase-type plasminogen activator and in-

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creasing release of plasminogen activator inhibitor-1 (PAI-1) [3–7].

At present, special attention is being paid to genetic predisposition to the severe coronavirus infection. Thus, it was shown in some studies that changes in the gene of angiotensin-converting enzyme 2 (ACE2) altering the structure of this receptor could either facilitate or complicate penetration into a cell for the virus. Some data specifically indicate that functional deficiency of the ApoE protein under SARS-CoV-2 can promote progression of the disease and development of some complications; some studies discuss influence of HLA1 genes on the COVID-19 clinical course [8–11].

It is noteworthy that hereditary thrombophilia and its role in creating elevated risks of the infection caused by the SARS-CoV-2 virus have not been given relevant attention. Previously, some studies reported that hereditary high-risk thrombophilias such as factor II (prothrombin) mutation, factor V Leiden mutation, as well as allele variants in the PAI-1 gene can remain hidden during the whole life but such triggers as an injury, sepsis, or pregnancy can make them manifest themselves through organ disorders, progressing thrombinemia, thrombosis in various locations, and multiple organ dysfunction syndrome (MODS) in the most severe cases [7, 12, 13]. It is noteworthy that results of molecular-genetic analysis aimed at revealing prothrombotic polymorphisms in homeostasis genes can influence intensity and duration of anti-thrombotic therapy [7, 14–16].

At present, available and frequently applied laboratory tests do not give an opportunity to objectively and reliably predict a risk and severity of thrombinemia under COVID-19 in genetically predisposed patients and, consequently, to select an optimal anti-thrombotic prevention therapy.

In this study, our aim was to analyze molecular-genetic markers of homeostasis in patients with the new coronavirus infection COVID-19 as a prognostic trigger of developing prothrombotic readiness.

Materials and methods. A prospective clinical and laboratory study was accomplished during the COVID-19 pandemic (April 2020 – May 2021) in the Regional Center for Anti-Thrombotic Therapy of the E.E. Volosevich's First Municipal Clinical Hospital (Arkhangelsk). One hundred patients with average to severe and severe COVID-19 participated in it.

We applied several criteria to include patients into the study: they had PCR-confirmed COVID-19; they were hospitalized in a specialized COVID-19 unit; they gave their voluntary informed consent to participate in the study; their age was above 18 years. Exclusion criteria were as follows: refusal to take part in the study; a patient being younger than 18 years. The study design was approved by the Local Ethics Committee of the Northern State Medical University (the Meeting Report No. 2/20 dated April 23, 2020).

Laboratory tests were accomplished in the Regional Center for Anti-Thrombotic Therapy of the E.E. Volosevich's First Municipal Clinical Hospital (Arkhangelsk). Plasma samples were taken three times: first, when a patient was admitted to an in-patient hospital prior to application of any anti-thrombotic therapy; second, on the 3^{rd} – 5^{th} day of hospitalization when an anti-thrombotic therapy with low-molecular-weight heparin (LMWH) was being applied; third, on the 9^{th} – 10^{th} day of hospitalization under the ongoing anti-thrombotic therapy with LMWH.

Prothrombotic status was estimated by molecular-genetic analysis of venous blood taken from the ulnar vein by venipuncture into a vacutainer with ETDA (ethylendiaminetetraacetic acid), a sample volume being 4.5 ml. We analyzed samples of genome DNA extracted from peripheral blood leucocytes. Genotyping of polymorphisms and homeostasis mutations was performed by using the polymerase chain reaction (PCR) with subsequent restriction analysis of its product.

To analyze coagulation indicators (vacutainers with sodium citrate), the obtained blood samples were centrifuged under 3000 rpm for 15 minutes. According to the temporary methodical guidelines, version 11, that were valid at the moment this study was being accomplished, we identified prothrombin time (PT), D-dimer, activated partial thromboplastin time

(APTT) and fibrinogen with Sysmex CS-2000i blood hemostasis analyzer (Sysmex, Japan) during 30 minutes since the moment blood was taken into a vacutainers[1].

In addition, to estimate intensity of thrombinemia, we identified thrombin kinetics indicators using Ceveron® alpha fully automated coagulation analyzer with TGA-module and reagents produced by Ceveron TGA High (Technoclone GmbH, Austria). We measured and analyzed lag-time (tLag), time necessary to reach thrombin peak (tPeak), peak thrombin level (Peak), and endogenous thrombin potential (AUC).

Statistical analysis was performed using SPSS Statistics, version 20.0, and MedCalc software package. Quantitative variables are given as *Me* (median) and 25, 75 percentiles; qualitative data are given as relative frequency and 95 % confidence interval for a fraction. Quantitative data in dependent samples were compared with the unpaired two-samples Wilcoxon test. The critical level of statistical significance (p) was taken as equal to 0.05. We applied correlation analysis techniques (the Pearson's linear correlation coefficient and Spearman's rank correlation coefficient) and regression analysis techniques (multiple linear regression and multiple logistic regression).

Results and discussion. First, we analyzed severity of the new coronavirus infection. The analysis revealed that the median age of patients participating in the research was 63 [31; 85] years, women accounted for 60 % of them, and the 4th degree severity of the coronavirus pneumonia was identified in 56 % of the analyzed cases (Figure 1). Hospitalized patients with community-acquired pneumonia had the diseases with 4th degree of severity (SD) according to CT data; hospital-acquired pneumonia developed in one third of the patients and the 4th SD prevailed among them as well.

Actual clinical practice clearly indicates that laboratory diagnostics of prothrombotic readiness (thrombinemia) has a crucial role both in pathogenesis and intensive therapy of the new coronavirus infection COVID-19. At present, several laboratory markers are recommended by scientific societies as laboratory indicators of COVID-19-associated coagulopathy and inflammation [17, 18]. We should remember that recommended conventional or routine hemostasiological tests such as prothrombin time, activated partial thromboplastin time, levels of D-dimer and fibrinogen are not able to identify the actual prothrombin readiness under the new coronavirus infection or to predict severity of a prothrombotic state in a given patient. Table 1 provides the results of routine laboratory homeostasis indicators in COVID-19 patients analyzed in dynamics.

D-dimer levels were established to grow statistically significantly by the 4th hospitalization day despite the applied anti-thrombotic therapy with LMWH; fibrinogen and ferritin levels went down only by the 10th day of therapy (Figures 2 and 3). It should be noted that the aforementioned laboratory tests are laboratory markers for diagnosing both hypercoagulation and systemic inflammation [19, 20] and that routine clotting tests (APTT, INR) did not have any diagnostic significance to identify thrombinemia.

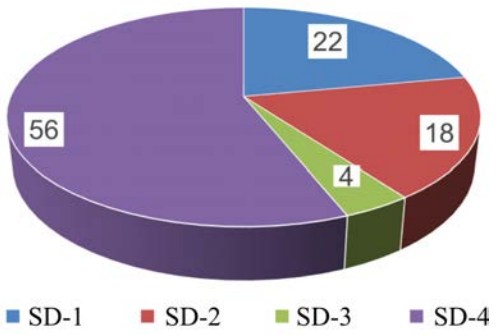


Figure 1. Degree of severity of the coronavirus infection according to lung computer tomography

[1] Profilaktika, diagnostika i lechenie novoi koronavirusnoi infektsii (COVID-19): Vremennye metodicheskie rekomendatsii. Versiya 11 (07.05.2021); utv. Zamestitelem Ministra zdravookhraneniya Rossiiskoi Federatsii E.G. Kamkinym [Prevention, diagnostics and treatment of the new coronavirus infection (COVID-19): Temporary methodical guidelines. Version 11 (May 07, 2021); approved by the Deputy to the RF Minister of Public Healthcare E.G. Kamkin]. *RF Public Healthcare Ministry*. Available at: http://nasci.ru/?id=40123&download=1 (March 30, 2022) (in Russian).

Table 1

Routine coagulogram indicators in COVID-19 taken in dynamics (Me [Q_1–Q_3])

Indicator	Hospitalization day		
	1st	4th	10th
Platelets, × 10^9/l	246 [85–407]	287 [70–615]	319 [179–500]*
APTT, sec	34.7 [30–47]	36.8 [30–49]	34.2 [23–79]
Fibrinogen, g/l	5.4 [2.3–6.8]	5.8 [3.2–6.5]	4.4 [2.8–7.2]**
D-dimer, mg/ml	1.16 [0.2–7.0]	1.6 [0.3–5.5]*	1.2 [0.1–4.2]**
INR, units	1.02 [0.8–1.0]	1.1 [0.9–1.1]	1.2 [1.0–1.2]

N o t e: *$p < 0.05$ means the difference from the 1st day is statistically significant; **$p < 0.05$ means the difference from the 4th day is statistically significant.

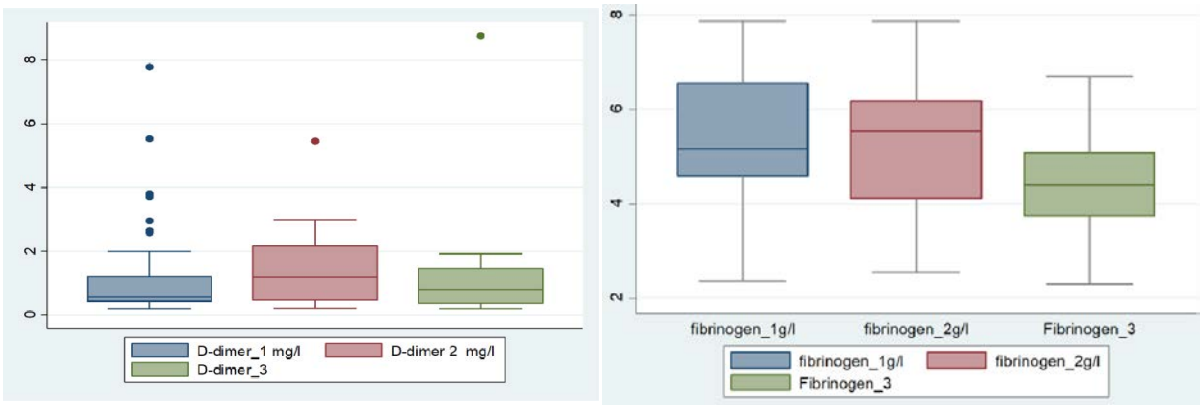


Figure 2. Levels of D-dimer (mg/l) and fibrinogen (g/l) taken in dynamics during hospitalization: the 1st, 4th, and the 10th day (n = 100)

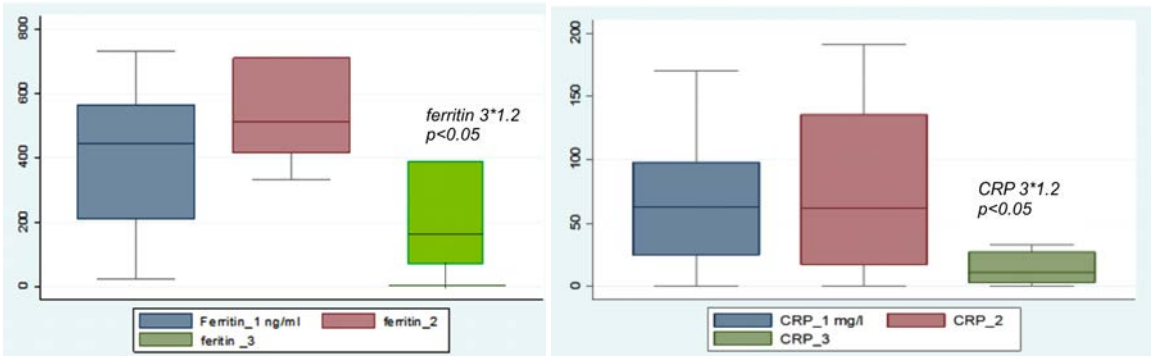


Figure 3. Levels of ferritin (ng/ml) and CRP (mg/l) taken in dynamics during hospitalization: the 1st, 4th, and the 10th day (n = 100)

We put forward a hypothesis that a hereditary genetically determined thrombophilic state could precede clinical signs of thrombinemia in some COVID-19 patients. This state involved an elevated risk of developing prothrombotic readiness against carriage of polymorphisms in fibrinogen, prothrombin and PAI-1 genes and its occurrence was associated with regulation of thrombin kinetics (generation). Given that, we believed it relevant to perform a molecular-genetic examination of COVID-19 patients to identify the occurrence of genetic polymorphisms in homeostasis indicating a genetically determined thrombophilic state.

According to the basic aim of our study, we performed molecular-genetic analysis of basic genes significant for homeostasis including several factors determining its coagu-

lation section (Factor V (rs6025), Factor II (rs1799963), Factor I (rs1800790), Factor VII (rs6046), Factor XIII A1 (rs5985)); platelet section (IGN A2 (rs1126643), IGN B3 (rs5918)), and fibrinolysis activity (PAI-1 (rs1799889)). They all were recommended for personified pharmacotherapy (Table 2) [21]. The accomplished molecular-genetic tests revealed the 'wild-type' being the most frequent genetic polymorphism as per all the examined genes significant for homeostasis in this patient group. The only differences were PAI-1 gene (rs1799889) where heterozygous and homozygous polymorphisms prevailed (49 % and 34 % of the cases accordingly) (Table 2).

The next stage in the research involved analyzing associations between genetic polymorphisms of homeostasis factors (FII 20210 G>A (rs1799963), FV 1691 G>A (rs6025), PAI-1 675 5G>4G (rs1799889)) and severity of COVID-19-associated coagulopathy by using recommended routine tests. The analysis revealed a statistically significant growth in fibrinogen levels by the 4th day (more than 6.0 g/l; $p < 0.001$) in carriers of the heterozygous allele variant in PAI-1, factor V and II genes. D-dimer levels changed as well but this change was not authentic; they tended to grow in most patients regardless of a genotype of analyzed homeostasis factors (Table 3).

According to the International Statistical Classification of Diseases and Related Health Problems, 10th Revision (ICD-10), mutations of factor II and V genes are among hereditary high-risk thrombophilias. Given that, we investigated possible associations between genetic polymorphisms of prothrombin FII 20210 G>A and V Leiden G>A genes and intensity of thrombinemia in COVID-19 patients. Our analysis revealed that thrombinemia was more intense in patients with genetic polymorphisms of the said genes (Figure 4).

Table 2

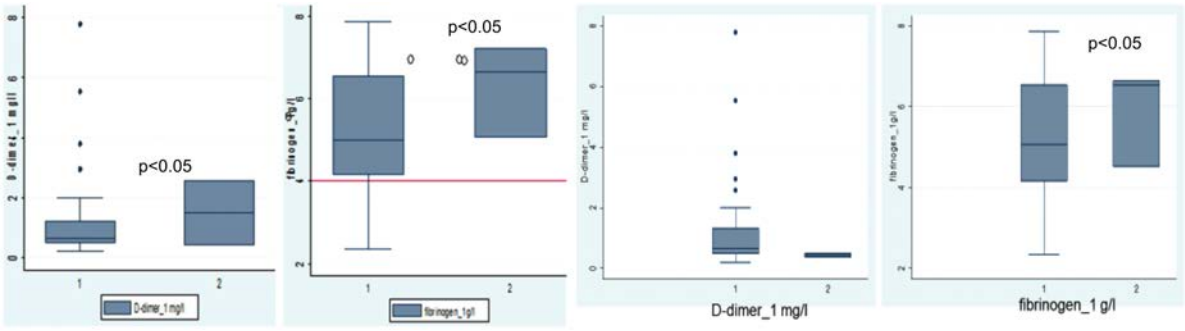
Genetic polymorphisms of homeostasis genes in COVID-19 patients ($n = 100$)

Analyzed genes	Genotype	Frequency, %	95 % CI
Factor XIII A1 (rs5985)	103 (G/G)	47.0	41.0; 53.8
	103 (G/T)	40.0	33.7; 46.0
	103 (T/T)	12.0	10.0; 15.4
IGN A2 (rs1126643)	807 (C/C)	43.0	45.0; 59.0
	807 (C/T)	38.0	31.5; 44.0
	807 (T/T)	19.0	17.0; 22.4
IGN B3 (rs5918)	1565(T/T)	66.0	60.0; 73.0
	1565(T/C)	26.0	19.0; 31.0
	1565(C/C)	12.0	10.0; 15.4
Factor V (rs6025)	1691 (G/G)	91.0	89.2; 92.8
	1691 (G/A)	6.0	4.2; 7.8
	1691 (A/A)	0	-
Factor II (rs1799963)	20210 (G/G)	93.0	90.6; 94.0
	20210 (G/A)	2.0	1.2; 4.4
	20210 (A/A)	0	-
PAI-1 (rs1799889)	–675 5G/5G	17.0	11.95; 22.51
	–675 4G/5G	49.0	42.2; 56.1
	675 4G/4G	34.0	30.8; 40.8
Factor I (rs1800790)	455 G/G	57.0	51.0; 63.8
	–455 G/A	37.0	30.7; 42.9
	–455 A/A	6.0	3.7; 9.4
Factor (VII rs6046)	10976 G/G	72.0	65.1; 78.2
	10976 G/A	23.0	17.0; 29.8
	10976 A/A	5.0	3.5; 7.9

Table 3

Thrombinemia intensity depending on allele variants in PAI-1, II and V factor genes

Genetic polymorphism	Hospitalization day					
	1st	4th	10th	1st	4th	10th
	D-dimer, mg/l (*Me*)			Fibrinogen, g/l (*Me*)		
PAI-I675 5G>4G (rs1799889)						
5G/5G (*n* = 17)	0.9	1.5	3.2	4.9	4.6	4.0
4G/5G (*n* = 49)	1.4	1.5	1.5	5.5	5.0	4.6
4G/4G (*n* = 34)	0.9	0.9	0.7	5.3	5.7	4.1
FV 1691 G>A (rs6025)						
GG (*n* = 91)	1.3	1.8	1.2	5.4	5.0	4.5
GA (*n* = 6)	0.7	0.5	1.1	5.9	6.7	3.7
FII 20210 G>A (rs1799963)						
GG (*n* = 93)	0.6	1.5	3.03	5.0	5.15	4.4
GA (*n* = 2)	2.6	0.2	0.4	6.0	6.7	4.1



1 – standard; *2* – heterozygous polymorphism

1 – standard; *2* – heterozygous polymorphism

Figure 4. Levels of D-dimer and fibrinogen upon admission to hospital in patients with polymorphisms of FII 20210 G>A and FV 1691 G>A genes

We performed regression analysis of independent thrombinemia predictors and a dependent variable (D-dimer level upon admission, the 1st day). The analysis revealed that a risk of a D-dimer level growing higher than 0.5 mg/l increased for patients with genetic polymorphisms of the PAI-1 gene (the heterozygous allele variant, β 95 % CI: 1.4 [0.6–2.13], p = 0.001; the homozygous allele variant, β 95 % CI: 2.0 [0.3–1.5], p = 0.008) and the heterozygous polymorphism of factor II (prothrombin) gene (Table 4).

According to multifactor analysis data, genetic polymorphism of the PAI-1 gene and ferritin levels higher than 200 pg/ml had authentic influence on an increase in D-dimer levels above their reference range (Table 5).

To achieve objectivity in assessing thrombinemia by laboratory tests, we applied a thrombin generation (kinetics) test (TGT). It shows how much thrombin is generated and kinetics of its generation thereby assessing the state of prothrombotic readiness [22, 23]. The results of the thrombin generation tests which we obtained in this study indicated an increase in pro-coagulation blood potential in COVID-19 patients already upon admission to in-patient hospital. Thus, we established that all the thrombin kinetics indicators changed statistically significantly in patients on the 1st day of hospitalization. This indicated significant activation of thrombin and occurring thrombinemia, or prothrombotic readiness (Table 6).

Table 4

Linear regression (D-dimer and genetic polymorphism)

Independent predictors	D-dimer		
	β	*p*	95 % CI
FV (rs6025) wild-type	1.2	0.1	-2.4; 2.27
-heterozygous polymorphism	0.7	0.2	0.1; 1.1
-homozygous polymorphism	1.0	0.3	0.31; 1.5
PAI-1 (rs1799889) wild-type	0.9	0.1	-0.43; 1.2
- heterozygous polymorphism	1.4	0.001	0.61; 2.13
- homozygous polymorphism	2.0	0.008	0.31; 1.5
FII (rs1799963) wild-type	1.0	0.6	
- heterozygous polymorphism	0.3	0.01	0.3; 1.9

Table 5

Regression analysis of independent thrombinemia predictors and D-dimer dependent variable upon admission (the 1st day)

Thrombinemia predictors	OR [95 % CI]	*p*
Genetic polymorphism in PAI-1 gene (rs1799889)	1.2 [0.1–2.5]	0.005
Ferritin level higher than 200 pg/ml	2.4 [1.1–5.4]	0.036
CRP level higher than 5.0 mg/l	0.1 [0.01−0.7]	0.999

Table 6

Thrombin generation test indicators in COVID-19 patients at the moment they were included into the study and prior to any anti-coagulant therapy

Analyzed indicator	*Me* [Q_1–Q_3]	Reference value
International normalized ratio (Tlag), min	2.46 [1.3–4.2]	7.8–13.6
Endogenous prothrombin potential (AUC, nM), nMol/min	4425.1 [3400–5070]	1379.4–1735.9
Peak thrombin level in a sample (Peak), nMol/min	862.43 [680.4–1040]	98.4–153.7
Time necessary to reach the peak (tPeak), min	5.87 [4.4–7.5]	16.7–23.2

The final stage in our study involved analyzing an association between polymorphisms of homeostasis genes and the thrombin generation test as a prognostic trigger indicating a risk of developing prothrombotic readiness in COVID-19 patients. The correlation analysis of the international normalized ratio (Tlag) and factor I (fibrinogen) genotypes revealed a moderate negative correlation; that is, a heterozygous polymorphism of the fibrinogen gene is associated with decreasing time of a lag in blood clotting. Both heterozygous and homozygous polymorphisms in the fibrinogen gene were associated with a growing peak thrombin level (Peak), which was authentically higher than in COVID-19 patients without such genetic polymorphisms (Figure 5).

Both heterozygous and homozygous polymorphisms of the PAI-1 gene were associated with shorter time necessary to reach the peak thrombin level (tPeak); that is, the peak thrombin level was reached faster in patients with alternative polymorphism of the PAI-I gene (Figure 6).

Correlation analysis of associations between levels of endogenous thrombin potential (AUC) and genotypes of the clotting factor genes I, II and PAI-1 indicated that alternative polymorphisms of the fibrinogen and PAI-1 genes were associated with elevated endogenous thrombin potential (Figure 7).

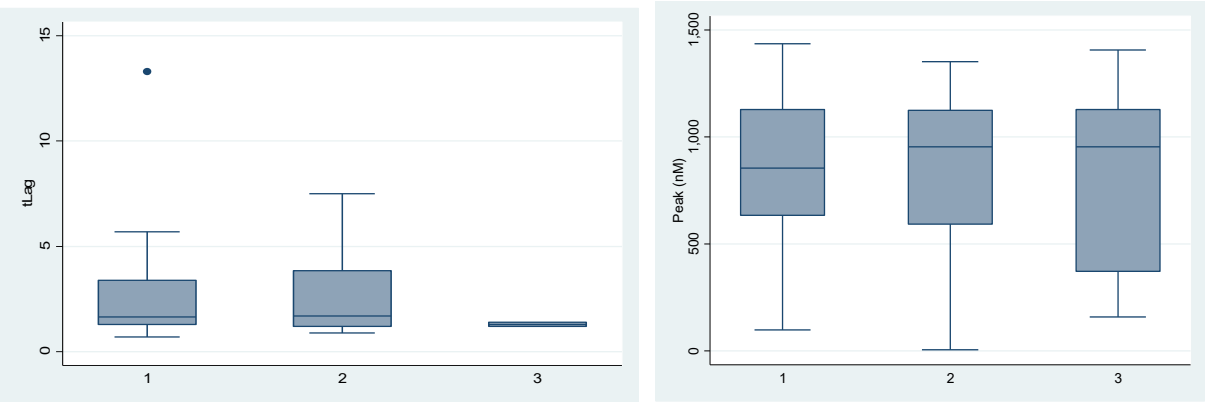


Figure 5. Correlation analysis between international normalized ratio (tLag), peak thrombin level (Peak thrombin, nmol/l) and factor I clotting genotypes (fibrinogen)

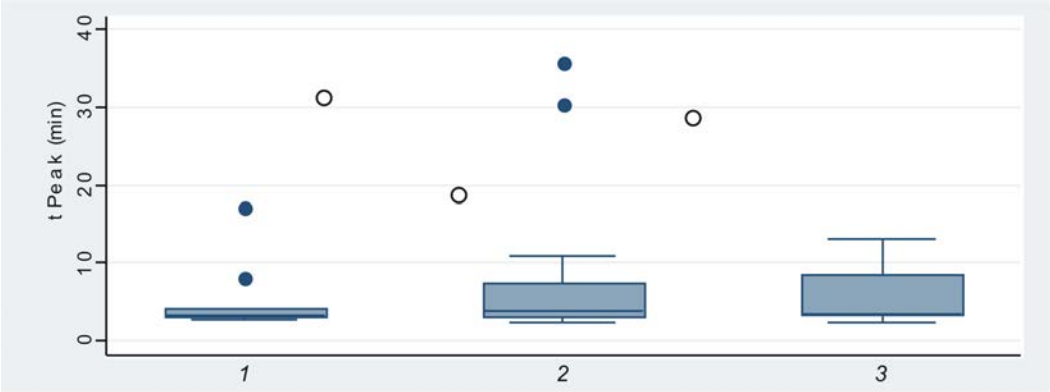


Figure 6. Analysis of correlations between time necessary to reach peak thrombin level (tPeak, min) and genetic polymorphisms of the PAI-1 gene: 1 is wild-type, 2 is heterozygous allele variant, 3 is homozygous allele variant

Figure 7. Analysis of correlations between levels of endogenous thrombin potential (AUC) and genotypes of the PAI-1, clotting factors II and I genes: 1 is wild-type, 2 is heterozygous allele variant, 3 is homozygous allele variant

Conclusion. Therefore, polymorphisms of PAI-1, prothrombin (FII), and fibrinogen (FI) genes determined high thrombinemia according to thrombin generation test indicators (endogenous thrombin potential (AUC), peak thrombin level (Peak thrombin), time necessary to reach the peak thrombin level (tPeak), fibrinogen and D-dimer levels) in COVID-19 patients over the whole hospitalization period. The study results indicate that prothrombotic

readiness is a genetically determined state in COVID-19 patients with allele variants in the PAI-1, prothrombin (factor II) and fibrinogen (factor I) genes. Elevated thrombin generation that became apparent through elevated endogenous thrombin potential (AUC) was shown to be a possible prognostic sign of prothrombotic readiness in patients with genetic polymorphisms of the PAI-1 and fibrinogen genes.

The accomplished pilot study showed that molecular-genetic testing aimed at identifying hereditary-determined thrombinemia could be considered a prognostic marker indicating an existing risk of developing prothrombotic readiness in patients with average to severe and severe COVID-19. The data obtained by this prospective clinical investigation prove the suggested molecular-genetic thrombinemia screening to be useful in treating COVID-19 patients. A clinician in a 'red zone' of a COVID-19 hospital is able to rely on an additional objective indicator that predicts thrombinemia in COVID-19 patients with acute inflammation.

Screening of genetic polymorphisms in medicine dealing with critical conditions is vital when a patient is given a pathogenetically justified antithrombotic therapy as well as within prevention activities. Our study results indicate that thrombinemia in COVID-19 patients is likely to have certain molecular mechanisms and that it is advisable to widely implement DNA-diagnostics into clinical practice for assessing severity of prothrombotic readiness and predicting its development.

Following the results of this study, a patent for invention No. 2789822 was issued on February 02, 2023.

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Competing interests. The authors declare no manifest or potential competing interests related to publishing this research article.

References

1. Katsoularis I., Fonseca-Rodríguez O., Farrington P., Jerndal H., Häggström Lundevaller E., Sund M., Lindmark K., Fors Connolly A.-M. Risks of deep vein thrombosis, pulmonary embolism, and bleeding after COVID-19: nationwide self-controlled cases series and matched cohort study. *BMJ*, 2022, vol. 377, pp. e069590. DOI: 10.1136/bmj-2021-069590

2. Bikdeli B., Madhavan M.V., Jimenez D., Chuich T., Dreyfus I., Driggin E., Der Nigoghossian C., Ageno W. [et al.]. COVID-19 and Thrombotic or Thromboembolic Disease: Implications for Prevention, Antithrombotic Therapy, and Follow-up: JACC State-of-the-Art Review. *J. Am. Coll. Cardiol.*, 2020, vol. 75, no. 23, pp. 2950–2973. DOI: 10.1016/j.jacc.2020.04.031

3. Wu C., Chen X., Cai Y., Xia J., Zhou X., Xu S., Huang H., Zhang L. [et al.]. Risk factors associated with acute respiratory distress syndrome and death in patients with coronavirus disease 2019 pneumonia in Wuhan, China. *JAMA Intern. Med.*, 2020, vol. 180, no. 7, pp. 934–943. DOI: 10.1001/jamainternmed.2020.0994

4. Soares M.P., Teixeira L., Moita L.F. Disease tolerance and immunity in host protection against infection. *Nat. Rev. Immunol.*, 2017, vol. 17, no. 2, pp. 83–96. DOI: 10.1038/nri.2016.136

5. Kabouridis P.S., Jury E.C. Lipid rafts and T-lymphocyte function: implications for autoimmunity. *FEBS Lett.*, 2008, vol. 582, no. 27, pp. 3711–3718. DOI: 10.1016/j.febslet.2008.10.006

6. Mannucci P.M., Franchini M. Classic thrombophilic gene variants. *Thromb. Haemost.*, 2015, vol. 114, no. 5, pp. 885–889. DOI: 10.1160/TH15-02-0141

7. Haralambous E., Hibberd M.L., Hermans P.W., Ninis N., Nadel S., Levin M. Role of functional plasminogen-activator-inhibitor-1 4G/5G promoter polymorphism in susceptibility, severity, and outcome of meningococcal disease in Caucasian children. *Crit. Care Med.*, 2003, vol. 31, no. 12, pp. 2788–2793. DOI: 10.1097/01.CCM.0000100122.57249.5D

8. Wan Y., Shang J., Graham R., Baric R.S., Li F. Receptor recognition by the Novel Coronavirus from Wuhan: an analysis based on decade-long structural studies of SARS Coronavirus. *J. Virol.*, 2020, vol. 94, no. 7, pp. e00127-20. DOI: 10.1128/JVI.00127-20

9. Varga Z., Flammer A.J., Steiger P., Haberecker M., Andermatt R., Zinkernagel A.S., Mehra M.R., Schuepbach R.A. [et al.]. Endothelial cell infection and endotheliitis in COVID-19. *Lancet*, 2020, vol. 395, no. 10234, pp. 1417–1418. DOI: 10.1016/S0140-6736(20)30937-5

10. Moore J.B., June C.H. Cytokine release syndrome in severe COVID-19. *Science*, 2020, vol. 368, no. 6490, pp. 473–474. DOI: 10.1126/science.abb8925

11. Huang C., Wang Y., Li X., Ren L., Zhao J., Hu Y., Zhang L., Fan G. [et al.]. Clinical features of patients infected with 2019 novel coronavirus in Wuhan, China. *Lancet*, 2020, vol. 395, no. 10223, pp. 497–506. DOI: 10.1016/S0140-6736(20)30183-5

12. Vorobyova N.A., Nedashkovsky E.V. Optimization of intensive care in acute disseminated intravascular syndrome. *Anesteziologiya i reanimatologiya*, 2003, no. 4, pp. 50–54 (in Russian).

13. Vorob'eva N.A. Mesto geneticheskikh polimorfizmov sistemy gemostaza v geneze trombofilicheskikh sostoyanii [Role of genetic polymorphisms of the hemostasis system in the genesis of thrombophilic conditions]. *Arctic environmental research*, 2004, no. 2 (6), pp. 14–21 (in Russian).

14. Garred P., Strom J.J., Quist L., Taaning E., Madsen H.O. Association of mannose-binding lectin polymorphisms with sepsis and fatal outcome, in patients with systemic inflammatory response syndrome. *J. Infect. Dis.*, 2003, vol. 188, no. 9, pp. 1394–1403. DOI: 10.1086/379044

15. Gordon A.C., Lagan A.L., Aganna E., Cheung L., Peters C.J., McDermott M.F., Millo J.L., Welsh K.I. [et al.]. TNF and TNFR polymorphisms in severe sepsis and septic shock: a prospective multicentre study. *Genes Immun.*, 2004, vol. 5, no. 8, pp. 631–640. DOI: 10.1038/sj.gene.6364136

16. Vorobyova N.A., Kapustin S.I. Role of hemostatic system's genetic monitoring during serious proceeding of acute syndrome of disseminated intravascular coagulation. *Ekologiya cheloveka*, 2005, no. 12, pp. 25–30 (in Russian).

17. The International Society on Thrombosis and Hemostasis (ISTH) interim guidance on recognition and management of coagulopathy in COVID-19: digest. *Aterotromboz*, 2020, no. 1, pp. 6–8. DOI: 10.21518/2307-1109-2020-1-6-8 (in Russian).

18. Shatohin Yu.V., Snezhko I.V., Ryabikina E.V. Violation of hemostasis in coronavirus infection. *Yuzhno-Rossiiskii zhurnal terapevticheskoi praktiki*, 2021, vol. 2, no. 2, pp. 6–15. DOI: 10.21886/2712-8156-2021-2-2-6-15 (in Russian).

19. Linkins L.A., Takach Lapner S. Review of D-dimer testing: good, bad, and ugly. *Int. J. Lab. Hematol.*, 2017, vol. 39, suppl. 1, pp. 98–103. DOI: 10.1111/ijlh.12665

20. Thachil J., Lippi G., Favaloro E.J. D-dimer testing: laboratory aspects and current issues. *Methods Mol. Biol.*, 2017, vol. 1646, pp. 91–104. DOI: 10.1007/978-1-4939-7196-1_7

21. Colucci G., Tsakiris D.A. Thrombophilia screening revisited: an issue of personalized medicine. *J. Thromb. Thrombolysis*, 2020, vol. 49, no. 4, pp. 618–629. DOI: 10.1007/s11239-020-02090-y

22. Hemker H.C., Al Dieri R., De Smedt E., Béguin S. Thrombin generation, a function test of the haemostatic-thrombotic system. *Thromb. Haemost.*, 2006, vol. 96, no. 5, pp. 553–561.

23. Carlier L., Hunault G., Lerolle N., Macchi L. Ex vivo thrombin generation patterns in septic patients with and without disseminated intravascular coagulation. *Thromb. Res.*, 2015, vol. 135, no. 1, pp. 192–197. DOI: 10.1016/j.thromres.2014.11.001

Vorobyeva N.A., Vorobyeva A.I., Vorontsova A.S. Predicting risks of prothrombotic readiness under COVID-19 using genetic testing. Health Risk Analysis, 2023, no. 2, pp. 130–139. DOI: 10.21668/health.risk/2023.2.12.eng

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