Journal of the American Heart Association

ORIGINAL RESEARCH

Prothrombin G20210A and Factor V Leiden Variants Are Not Associated With Thrombotic Events in Congenital Heart Disease: An Observational Trial

Feria A. Ladha, MD, PhD; Christina VanderPluym, MD; Enrique Mondragon-Estrada, BS; Paul Avillach, MD, PhD; Martina Brueckner, MD; Wendy K. Chung, MD, PhD; James F. Cnota, MD; Bruce D. Gelb, MD; Matthew Lewis, MD; Cong Liu, PhD; Amy E. Roberts, MD; Christine E. Seidman, MD; Martin Tristani-Firouzi, MD; Michael Wagner, PhD; Sarah U. Morton, MD, PhD*; Jane W. Newburger, MD*; on behalf of the Pediatric Cardiac Genomics Consortium Investigators†

BACKGROUND: Thrombotic events, including acute ischemic stroke, are more common in individuals with congenital heart disease (CHD). Whether common thrombophilia variants contribute to thrombotic risk in this population remains unclear. We evaluated whether prothrombin G20210A (*F2* c.97G>A) and factor V Leiden (*F5* c.1601G>A; p.Arg534Gln) are associated with thrombotic events in CHD.

METHODS: Participants in the Pediatric Cardiac Genomics Consortium with exome sequencing and electronic medical record data were identified. Individuals were stratified by prothrombin G20210A and factor V Leiden genotypes, ventricular physiology, and antithrombotic therapy. The primary outcome was the presence of *International Classification of Diseases* (*ICD*) or Phecodes (phenotype codes) for thrombotic events.

RESULTS: Among 4008 participants (median age, 11.4 [interquartile range, 5.1–17.9] years; 44.4% boys), thrombotic events occurred in 737 (18%), including 93 (13%) with acute ischemic stroke. Compared with the Genome Aggregation Database, the CHD cohort had a lower prevalence of heterozygous prothrombin G20210A and factor V Leiden variants. Variant prevalence did not differ between participants with and without thrombotic events. Single-ventricle CHD was associated with higher thrombosis frequency than biventricular CHD (35% versus 16%, $P \leq 0.0001$), without differences in variant prevalence.

CONCLUSIONS: In this multicenter CHD cohort, prothrombin G20210A and factor V Leiden were not significantly associated with thrombotic outcomes, supporting recommendations against routine screening. Given low variant prevalence, the study was powered to exclude only large associations. Reduced variant frequency suggests survivorship bias beginning in fetal life. Larger integrated clinical–genomic studies are needed to refine thrombotic risk stratification in CHD.

REGISTRATION: URL: https://www.clinicaltrials.gov; Unique Identifier: NCT03347214.

Key Words: congenital heart disease ■ factor V Leiden ■ stroke ■ thrombophilia ■ thrombosis

Correspondence to: Sarah U. Morton, MD, PhD, Boston Children's Hospital, 300 Longwood Ave, Boston, MA 02115. Email: sarah.morton@childrens.harvard.edu

*S. U. Morton and J. W. Newburger contributed equally.

†A complete list of the Pediatric Cardiac Genomics Consortium Investigators can be found in the Supplemental Material.

This manuscript was sent to Daniel E. Clark, MD, MPH, Associate Editor, for review by expert referees, editorial decision, and final disposition.

Supplemental Material is available at https://www.ahajournals.org/doi/suppl/10.1161/JAHA.125.046959

For Sources of Funding and Disclosures, see page 5.

JAHA is available at: www.ahajournals.org/journal/jaha

CLINICAL PERSPECTIVE

What Is New?

- In this large multicenter congenital heart disease (CHD) cohort, prothrombin G20210A and factor V Leiden variants were not associated with thrombotic events or acute ischemic stroke. The prevalence of these thrombophilia variants was lower in individuals with CHD than in the general population, suggesting possible survivorship bias beginning prenatally.
- Across clinical subgroups, including single-ventricle and biventricular CHD, variant carriers did not demonstrate increased thrombotic risk.

What Are the Clinical Implications?

- These findings support that routine screening for prothrombin G20210A or factor V Leiden should not guide thromboprophylaxis in individuals with CHD. Thrombotic risk stratification in CHD should continue to rely on clinical factors such as anatomy, hemodynamics, and surgical history rather than common thrombophilia genotypes. Future studies incorporating larger ancestrally diverse cohorts and integrated clinical–genomic models will be needed to refine individualized thrombotic risk prediction.

Nonstandard Abbreviation and Acronym

PCGC	Pediatric Cardiac Genomics Consortium

Congenital heart disease (CHD) is the most common congenital malformation and affects ≈1% of live births.[1] Thrombotic events are an important complication, prompting anticoagulation with or without antiplatelet therapies for both thromboprophylaxis and treatment in this population.[2,3] Children with single-ventricle heart disease have an exceptionally high risk of thrombotic events including strokes.[4] By age 6 years, 20% of children with hypoplastic left heart syndrome or related single right ventricle disorders in the Single Ventricle Reconstruction prospective trial had at least 1 thrombotic event, and 7.5% had a stroke.[5] Indeed, approximately one-third of all pediatric strokes occur in children with CHD.[6] Other factors, including right-to-left intracardiac shunting, arrhythmias, prothrombotic state, cardiac surgery, heart failure, and systemic illness, have also been shown to increase risk of stroke among people with CHD.[7]

Genetic variants including those in *F2* (ie, prothrombin G20210A; rs1799963; *F2* c.97G>A) and *F5* (ie, factor V Leiden; rs6025; *F5* c.1601G>A; p.Arg534Gln) have been associated with an increased risk of intravascular thrombosis, thromboembolism, and cerebral stroke.[8] Although thrombophilia variants increase the relative risk of thromboembolic events in the general population,[9] their contribution to thrombotic events in people with CHD is unclear. We sought to determine whether common thrombophilia variants contributed to an increase in the risk of thrombotic events, including acute ischemic stroke in people with CHD.

METHODS

Deidentified individual participant data (including data dictionaries), study protocols, the statistical analysis plan, and the informed consent form will be made available upon publication to researchers who provide a methodologically sound proposal for use in achieving the goals of the approved proposal. Proposals should be submitted to b2bprogram@cchmc.org. Participants in the Pediatric Cardiac Genomics Consortium (PCGC)[10] with exome sequencing and electronic medical record (EMR) data were identified. The PCGC is a multicenter, prospective cohort established through the National Heart, Lung, and Blood Institute Bench to Bassinet program and includes people of all ages with CHD recruited from multiple pediatric cardiac centers across the United States. To minimize bias, clinical data are derived from standardized chart abstraction and harmonized EMR fields, including cardiac diagnoses, surgical history, and relevant comorbid conditions. Detailed phenotyping is supplemented by exome sequencing and biospecimen collection, enabling integration of genomic and clinical data. Enrollment started in December 2012. Written informed consent was obtained from participants or their parents. The study was approved by the institutional review board at each study center and, after 2023, by a single institutional review board. This study followed the Strengthening the Reporting of Observational Studies in Epidemiology reporting guideline. Thrombosis events were identified using PheCodes (phenotype codes; derived from *International Classification of Diseases*, *Ninth Revision* [*ICD-9*] and *Tenth Revision* [*ICD-10*] codes) 286.x, 411.x, 415.x, 433.x, 440.2, 444.x, 452.x, 586.3, 656.5, 671, 854, 856, 857, 858, and 859. PheCode 433.x was used to define acute ischemic stroke. Single-ventricle heart disease was identified on the basis of Fyler codes[1,2] 200, 210, 211, 213, 220, 230, 240, 250, 260, 270, 280, 300, 400, 411, 412, 413, 421, 422, 423, 431, 432, and 433. Antiplatelet agent (acetylsalicylic acid, clopidogrel, dipyridamole) or anticoagulation (apixaban, argatroban, bivalirudin, dabigatran etexilate, dalteparin, enoxaparin, fondaparinux, heparin, rivaroxaban, warfarin) use was identified using RxNorm codes. Central venous catheter use was identified using Current Procedural Terminology codes 36555, 36556, 36558,

Table 1. Thrombophilia Genotype Prevalence

	Prothrombin G20210A heterozygous, n (%)	OR (95% CI); *P* value	Factor V Leiden heterozygous, n (%)	OR (95% CI); *P* value
Genome Aggregation Database	16578/740 009 (2.2)	0.7 (0.5–0.8); <0.001	33674/80 658 (4.2)	0.6 (0.5–0.7); <0.001
PCGC	95/6371 (1.5)	…	158/6371 (2.5)	…
PCGC with electronic medical record data	53/4008 (1.3)	0.7 (0.5–1.1); 0.16	106/4008 (2.6)	1.2 (0.9–1.7); 0.28
PCGC without electronic medical record data	42/2363 (1.8)	…	52/2363 (2.2)	…

Prevalence of heterozygous *F2* and *F5* thrombophilia genotypes was compared for the following pairs: PCGC and Genome Aggregation Database, and with and without electronic medical record data among participants from PCGC. *P* value threshold of 0.0125 was used to define significance on the basis of Bonferroni correction for 4 test. OR indicates odds ratio; and PCGC, Pediatric Cardiac Genomics Consortium.

36573, and 36581. Exome data from PCGC and the Genome Aggregation Database[11] version 4.1 were subset to *F2* c.97G>A and *F5* c.1601G>A (hg38 chr11:46739505 and chr1:169549811, respectively) and filtered for quality as previously described.[3] Participants with missing data were excluded from analysis. Pearson's χ^2 test was used for comparing categorical outcomes. Power analyses were performed using the noncarrier event rates as baseline (18.4% for *F2*, 18.3% for *F5*) to calculate the minimum detectable effect size at 80% power (α=0.05), which corresponded to carrier event rates of ≥35% for *F2* (risk ratio, 1.9) and ≥29% for *F5* (risk ratio, 1.6).

RESULTS

Of 6371 PCGC participants with genotype data for thrombophilia variants prothrombin G20210A and factor V Leiden, 4008 also had EMR data (Table 1; Figure S1). Compared with the Genome Aggregation Database reference population, European participants in the PCGC cohort had a lower prevalence of heterozygous prothrombin G20210A and factor V Leiden variants (72/7634 [0.9%] versus 13811/535449 [2.6%], *P*<0.001; and 126/7634 [1.7%] versus 29147/589584 [5%], *P*<0.001, respectively). The frequency did not differ for participants with predominantly African (*F2*: 3/568 [0.53%] versus 165/36334 [0.45%], *P*=0.75; *F5*: 7/568 [1.2%] versus 282/37506 [0.75%], *P*=0.21), Admixed American (*F2*: 18/1421 [1.27%] versus 671/29247 [2.3%], *P*=0.02; *F5*: 20/1421 [1.4%] versus 460/30013 [1.5%], *P*=0.82), or South Asian (*F2*: 2/362 [0.55%] versus 173/43590 [0.4%], *P*=0.66; *F5*: 5/362 [1.4%] versus 1270/45534 [2.8%], *P*=0.14) genetic ancestry. In the overall cohort, the prevalence of homozygous variants did not differ significantly, with limited statistical power for comparison. Nine individuals (0.1%) were compound heterozygotes, carrying both *F2* and *F5* risk alleles.

Among the 4008 PCGC participants with EMR data, 737 (18%) had PheCodes for a peripheral or central thrombotic event; these included acute ischemic stroke (n=93/737 [13%]) and other events such as peripheral venous thrombosis or cerebral embolism (n=644/737 [87%]). Central venous catheter exposure was present in 689 of the 4008 participants (17%), and its prevalence did not differ between those with and without thrombotic events (112/737 [15%] versus 577/3271 [18%], *P*=0.12). At least 1 antithrombotic medication, including antiplatelet agents and anticoagulants, was prescribed at some point to 1417 of the 4008 PCGC participants (37%).

Participants with single-ventricle heart disease, compared with biventricular heart disease, had a higher frequency of thrombosis (166/520 [32%] versus 571/3488 [16%], *P*≤0.001) and were more likely to have received antithrombotic agents (Table 2). Thromboprophylaxis was administered for more participants with codes for thrombotic events than for those without such codes (384/459 [84%] versus 627/958 [65%], *P*≤0.001).

PCGC participants with and without EMR data did not differ significantly in thrombophilia variant prevalence (Table 3). Among those with EMR data, the prevalence of prothrombin G20210A and factor V Leiden variants did not differ significantly according to univentricular versus biventricular status. Heterozygous thrombophilia genotypes were found in 30 (4%) of the 737 participants with codes for thrombotic events. The prevalence of a thrombophilia variant did not differ significantly according to presence or absence of codes for a thrombotic event or an acute ischemic stroke. Among the 93 participants with acute ischemic stroke, 2 were prothrombin G20210A heterozygous, and 1 was factor V Leiden heterozygous. Six of the 9 individuals who were heterozygotes for both *F2* and *F5* risk alleles had EMR data, and 2 (25%) had thrombotic events.

DISCUSSION

In this multicenter CHD cohort, we found no significant association between heterozygous prothrombin G20210A or factor V Leiden variants and thrombotic outcomes in CHD. Only a small proportion of participants with thrombotic events carried 1 of these variants, and no enrichment was observed compared with those without events. These data suggest that,

Table 2. Thromboprophylaxis and Thrombosis Events

	None, n (%)	Antiplatelet agent, n (%)	Anticoagulation agent, n (%)	Any vs none, OR (95% CI); *P* value
Overall	406/1417 (28.7)	924/1417 (65.2)	319/1417 (22.5)	…
Single ventricle	53/243 (21.8)	163/243 (67.1)	80/243 (32.9)	1.5 (1.1–2.1); 0.012
Biventricular	353/1174 (30.1)	761/1174 (64.8)	239/1174 (20.4)	…
Thrombosis	75/459 (16.3)	341/459 (74.3)	189/459 (41.2)	2.7 (2.0–3.6); <0.001
No thrombosis	331/958 (34.6)	583/958 (60.9)	130/958 (13.6)	…

Prevalence of thromboprophylaxis in the overall cohort as well as by CHD type and thrombosis status. *P* value threshold of 0.025 was used to define significance on the basis of Bonferroni correction for 2 test. CHD indicates congenital heart disease; and OR, odds ratio.

in the era of routine prophylactic antithrombotic therapy, common thrombophilia variants contribute little to thrombotic risk in CHD. Instead, hemodynamic burden, intracardiac shunting, surgical interventions, and flow disturbances likely play a greater role in driving outcomes. Central venous catheter exposure, another known risk factor for thrombotic events, was not more frequent among those with thrombotic events in our cohort, although catheter-related risk may have been obscured by concurrent hemodynamic and procedural factors, as well as antithrombotic prophylaxis. This aligns with current clinical practice, which does not recommend routine screening for *F2* or *F5* variants when determining prophylactic therapy.

We also observed a lower prevalence of heterozygous prothrombin G20210A and factor V Leiden variants compared with the general population in people of European genetic ancestry. Prior studies reporting higher frequencies were often single-center or selectively recruited, differing demographically from ours and sometimes including patients without CHD or perioperative or high-risk subsets.[12,13] These findings suggest survivorship bias. Thrombophilia variants including factor V Leiden and prothrombin G20210A have been linked to placental thrombosis, fetal loss, and adverse perinatal outcomes, and carriers with concomitant CHD may face even higher risks of not surviving to enrollment.[14,15] Thus, the lower prevalence in our CHD cohort may reflect attrition beginning as early as fetal life. Larger, ancestrally diverse cohorts will be required to determine whether survivorship bias underlies the reduced prevalence of thrombophilia variants in CHD.

We relied on EMR and medication data from cardiac centers, which may contain coding errors or omissions, and thrombotic events occurring in outpatient or nonparticipating inpatient settings may not have been captured. The timing of antithrombotic therapy relative to events could not be determined. Even in hospitalized patients, diagnostic codes likely underestimate event frequency, as many thrombotic events, particularly cerebral infarcts, are clinically silent. Additionally, although prior studies report that many patients with single-ventricle CHD experience thrombosis by early childhood, we could not determine the precise timing of thrombotic events in our cohort due to limitations in EMR documentation, preventing direct comparison with age-specific rates reported in the literature. For example, brain magnetic resonance imaging studies after the Fontan procedure demonstrate that a substantial proportion of strokes go undiagnosed clinically.[16] Due to the low prevalence of prothrombin

Table 3. Thrombophilia Genotypes and Thrombosis

	Female/ Male sex	Age, y, median (IQR)	Prothrombin G20210A *F2* c.*97G>A heterozygous, n (%)	OR (95% CI); *P* value	Factor V Leiden heterozygous, n (%)	OR (95% CI); *P* value
Overall	2228/1780	11.4 (5.1–17.9)	53/4008 (1.3)	…	106/4008 (2.6)	…
Single-ventricle CHD	201/319	10.5 (5.1–17.5)	3/520 (0.6)	0.4 (0.1–1.2); 0.15	7/520 (1.4)	0.5 (0.2–1.0); 0.06
Biventricular CHD	1579/1909	11.5 (5.1–17.9)	50/3488 (1.4)	…	99/3488 (2.8)	…
Thrombosis	309/428	10.6 (5.6–18.0)	8/737 (1.1)	0.8 (0.3–1.7); 0.79	20/737 (2.7)	1.0 (0.6–1.7); 0.90
No thrombosis	1471/1800	11.5 (4.9–17.9)	45/3271 (1.4)	…	86/3271 (2.6)	…
Acute ischemic stroke	45/48	17.1 (7.1–31.0)	2/93 (2.2)	2.3 (0.2–13.3); 0.27	1/93 (1.1)	0.4 (0.0–2.3); 0.50
No acute ischemic stroke	264/380	10.1 (5.4–17.9)	6/644 (0.9)	…	19/644 (3.0)	…

Sex balance and age at last electronic medical data are provided for the overall cohort and each subset. Prevalence of *F2* and *F5* risk genotypes was compared for the following pairs: single-ventricle and biventricular CHD, with and without thrombosis, and with and without acute stroke among those with thrombosis. *P* value threshold of 0.0008 was used to define significance on the basis of Bonferroni correction for 4 tests. CHD indicates congenital heart disease; IQR, interquartile range; and OR, odds ratio.

G20210A or factor V Leiden variants, the study was powered only to detect large effects on thrombotic event rates (≥1.9-fold for *F2* and ≥1.6-fold for *F5*), and smaller relative increases could not be reliably excluded. Calculations of the minimum detectable effect size showed that only large relative risks could be detected and that additional sensitivity or subgroup analyses would have been underpowered and statistically unstable. Thrombophilia genotypes beyond prothrombin G20210A and factor V Leiden were not assessed, and our study was underpowered to evaluate contributions from rare homozygous or compound genotypes.

Taken together, our findings clarify 2 points: (1) that common thrombophilia variants appear reduced in prevalence in CHD, possibly reflecting survivorship bias; and (2) that these variants are not significantly associated with thrombotic outcomes in this population. Future work incorporating ancestry-stratified analyses; larger and more diverse cohorts; and models integrating clinical, procedural, and genomic factors will be essential to refine thrombotic risk prediction and optimize prophylaxis for individuals with CHD.

ARTICLE INFORMATION

Received October 3, 2025; accepted January 30, 2026.

Affiliations

Department of Pediatrics (F.A.L., W.K.C., A.E.R.), Department of Cardiology (C.V., A.E.R., J.W.N.), Division of Newborn Medicine, Department of Pediatrics (E.M.-E., S.U.M.), Fetal Neonatal Neuroimaging and Developmental Science Center (E.M.-E.) and Computational Health Informatics Program, Boston Children's Hospital, Boston, MA (P.A.); Department of Pediatrics, Harvard Medical School, Boston, MA (P.A., W.K.C., S.U.M., J.W.N.); Department of Epidemiology, Harvard T. H. Chan School of Public Health, Boston, MA (P.A.); Department of Genetics and Pediatrics, Yale University School of Medicine, New Haven, CT (M.B.); Department of Pediatrics and Medicine, Columbia University Medical Center, New York, NY (W.K.C.); Heart Institute, Cincinnati Children's Hospital Medical Center, Cincinnati, OH (J.F.C.); Mindich Child Health and Development Institute and Department of Pediatrics, Icahn School of Medicine at Mount Sinai, New York, NY (B.D.G.); Division of Cardiology, Department of Medicine, Columbia University, New York, NY (M.L.); Division of Genetics and Genomics, Department of Pediatrics (C.L.) and Division of Genetics, Department of Pediatrics, Boston Children's Hospital, Boston, MA (A.E.R., C.E.S.); Department of Cardiology, Brigham and Women's Hospital, Boston, MA (C.E.S.); Division of Pediatric Cardiology, University of Utah School of Medicine, Salt Lake City, UT (M.T.-F.); Divisions of Biomedical Informatics and of Biostatistics and Epidemiology, Cincinnati Children's Hospital Medical Center, Cincinnati, OH (M.W.); and Department of Pediatrics, University of Cincinnati College of Medicine, Cincinnati, OH (M.W.).

Acknowledgments

The authors thank all participants and their families.

Sources of Funding

This project was supported by the National Center for Advancing Translational Sciences (UL1-TR000003 and UL1-TR002541), the National Institutes of Health (U01-HL098188, U01-HL098147, U01-HL098153, U01-HL098163, U01-HL098123, U01-HL098162, U01-HL131003, 5R01-HL162356, 5U54-HG006504 [C.E.S. and S.U.M] and K08-HL157653 [S.U.M.]), Boston Children's Hospital Office of Faculty Development (Career Development Award, S.U.M.) and American Heart Association (Career Development Award, S.U.M.), the Farb Family Fund (J.W.N.), and the Kostin Family Innovation Fund (J.W.N.). The sponsors had no role in the design and conduct of the study.

Disclosures

None.

Supplemental Material

Figure S1
Pediatric Cardiac Genomics Consortium Investigators

REFERENCES

1. GBD 2017 Congenital Heart Disease Collaborators. Global, regional, and national burden of congenital heart disease, 1990–2017: a systematic analysis for the global burden of disease study 2017. *Lancet Child Adolesc Health*. 2020;4:185–200.
2. VanderPluym C, Esteso P, Ankola A, Hellinger A, Ventresco C, Hawkins B, Kobayashi RL, Williams R, Cetatoiu MA, Gauvreau K, et al. Real-world use of apixaban for the treatment and prevention of thrombosis in children with cardiac disease. *J Thromb Haemost*. 2023;21:1601–1609. doi: 10.1016/j.jtha.2023.03.005
3. Giglia TM, Massicotte MP, Tweddell JS, Barst RJ, Bauman M, Erickson CC, Feltes TF, Foster E, Hinoki K, Ichord RN, et al. Prevention and treatment of thrombosis in pediatric and congenital heart disease: a scientific statement from the American Heart Association. *Circulation*. 2013;128:2622–2703. doi: 10.1161/01.cir.0000436140.77832.7a
4. Van Den Helm S, Sparks CN, Ignjatovic V, Monagle P, Attard C. Increased risk for thromboembolism after Fontan surgery: considerations for thromboprophylaxis. *Front Pediatr*. 2022;10:803408.
5. Newburger JW, Sleeper LA, Gaynor JW, Hollenbeck-Pringle D, Frommelt PC, Li JS, Mahle WT, Williams IA, Atz AM, Burns KM, et al. Transplant-free survival and interventions at 6 years in the SVR trial. *Circulation*. 2018;137:2246–2253. doi: 10.1161/CIRCULATIONAHA.117.029375
6. Ferriero DM, Fullerton HJ, Bernard TJ, Billinghurst L, Daniels SR, DeBaun MR, deVeber G, Ichord RN, Jordan LC, Massicotte P, et al. Management of stroke in neonates and children: a scientific statement from the American Heart Association/American Stroke Association. *Stroke*. 2019;50:e51–e96. doi: 10.1161/STR.0000000000000183
7. Yelton SEG, Flores S, Sun LR, Nelson-McMillan K, Loomba RS. Association between congenital heart disease and stroke: insights from a national database. *Pediatr Cardiol*. 2024;45:1–7. doi: 10.1007/s00246-023-03315-6
8. Dautaj A, Krasi G, Bushati V, Precone V, Gheza M, Fioretti F, Sartori M, Costantini A, Benedetti S, Bertelli M, et al. Hereditary thrombophilia. *Acta Biomed*. 2019;90:44–46.
9. Crous-Bou M, Harrington LB, Kabrhel C. Environmental and genetic risk factors associated with venous thromboembolism. *Semin Thromb Hemost*. 2016;42:808–820.
10. Gelb B, Brueckner M, Chung WK, Goldmuntz E, Kaltman J, Kaski JP, Kim R, Kline J, Mercer-Rosa L, Porter G, et al. The congenital heart disease genetic network study: rationale, design, and early results. *Circ Res*. 2013;112:698–706. doi: 10.1161/CIRCRESAHA.111.300297
11. Chen S, Francioli LC, Goodrich JK, Collins RL, Kanai M, Wang Q, Alföldi J, Watts NA, Vittal C, Gauthier LD, et al. A genomic mutational constraint map using variation in 76,156 human genomes. *Nature*. 2024;625:92–100. doi: 10.1038/s41586-023-06045-0
12. Ong BC, Zimmerman AA, Zappulla DC, Neufeld EJ, Burrows FA. Prevalence of factor V Leiden in a population of patients with congenital heart disease. *Can J Anaesth*. 1998;45:1176–1180.
13. Ozbek N, Belgin Ataç F, Yildirim SV, Verdi H, Yazici C, Yilmaz BT, Tokel NK. Analysis of prothrombotic mutations and polymorphisms in children who developed thrombosis in the perioperative period of congenital cardiac surgery. *Cardiol Young*. 2005;15:19–25.
14. Meinardi JR, Middeldorp S, de Kam PJ, Koopman MM, van Pampus EC, Hamulyák K, Prins MH, Büller HR, van der Meer J. Increased risk for fetal loss in carriers of the factor V Leiden mutation. *Ann Intern Med*. 1999;130:736–739.
15. Gao H, Tao FB. Prothrombin G20210A mutation is associated with recurrent pregnancy loss: a systematic review and meta-analysis update. *Thromb Res*. 2015;135:339–346. doi: 10.1016/j.thromres.2014.12.001
16. Bellinger DC, Watson CG, Rivkin MJ, Robertson RL, Roberts AE, Stopp C, Dunbar-Masterson C, Bernson D, DeMaso DR, Wypij D, et al. Neuropsychological status and structural brain imaging in adolescents with single ventricle who underwent the Fontan procedure. *J Am Heart Assoc*. 2015;4:e002302. doi: 10.1161/JAHA.115.002302