Inherited thrombophilia and risk of thrombosis in children with cancer: a single-center experience

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Abstract

Background: Thrombosis is an increasingly recognized complication of childhood malignancy and its treatment. The etiology of pediatric cancer-related thrombosis is multifactorial and not well understood at present. The aim of this study was to evaluate the prevalence of common prothrombotic genetic conditions in children with cancer, the frequency of thrombosis, and the role of inherited thrombophilia in the development of thrombosis in a pediatric oncology population. Methods: Forty-seven children (36 treated for hematological malignancies and 11 for solid tumors) with the median age of 8.8. years (range 0.4 – 19.3 years) were included in the study. Genetic polymorphisms of Factor V Leiden, prothrombin G20210A mutation, and methylenetetrahydrofolate reductase (MTHFR) C677T were determined by real-time polymerase chain reaction-based DNA analysis. Results: Four (8.5%) patients were heterozygous for Factor V Leiden, 3 (6.4%) were heterozygous for prothrombin G20210A mutation, and 3 (6.4%) were homozygous for MTHFR C677T mutation. All patients had inserted central venous lines. Four (8.5%) children had documented thrombosis, 3 of which were located in the upper venous system. Two of four patients with thrombosis had Factor V Leiden heterozygosity. Conclusions: Thrombosis is an important complication of childhood cancer. Our results suggest that congenital prothrombotic abnormalities could be implicated in increasing the risk of thrombosis and support a recommendation that children with cancer be evaluated for inherited thrombophilia.

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Abbreviations

ALL	acute lymphoblastic leukemia
APCR	activated protein C resistance
CVL	central venous line
MTHFR	methylenetetrahydrofolate reductase
RT-PCR	real-time polymerase chain reaction

ABSTRACT

Background: Thrombosis is an increasingly recognized complication of childhood malignancy and its treatment. The etiology of pediatric cancer-related thrombosis is multifactorial and not well understood at present. The aim of this study was to evaluate the prevalence of common prothrombotic genetic conditions in children with cancer, the frequency of thrombosis, and the role of inherited thrombophilia in the development of thrombosis in a pediatric oncology population.

Methods: Forty-seven children (36 treated for hematological malignancies and 11 for solid tumors) with the median age of 8.8. years (range 0.4 – 19.3 years) were included in the study. Genetic polymorphisms of Factor V Leiden, prothrombin G20210A mutation, and methylenetetrahydrofolate reductase (MTHFR) C677T were determined by real-time polymerase chain reaction-based DNA analysis.

Results: Four (8.5%) patients were heterozygous for Factor V Leiden, 3 (6.4%) were heterozygous for prothrombin G20210A mutation, and 3 (6.4%) were homozygous for MTHFR C677T mutation. All patients had inserted central venous lines. Four (8.5%) children had documented thrombosis, 3 of which were located in the upper venous system. Two of four patients with thrombosis had Factor V Leiden heterozygosity.

Conclusions: Thrombosis is an important complication of childhood cancer. Our results suggest that congenital prothrombotic abnormalities could be implicated in increasing the risk of thrombosis and support a recommendation that children with cancer be evaluated for inherited thrombophilia.

INTRODUCTION

Thrombosis is a well-recognized complication of malignancy. It is estimated that up to 20% of all cancer patients develop thrombotic event during the course of the disease, with the annual incidence rate of 0.5% compared to 0.1% in the general population.[1,2] There is substantially less knowledge about thrombosis in pediatric cancer populations, with reported rates varying from 2 to 16%, depending on the type of

malignancy.[3] Children with cancer and thrombosis have increased risk of mortality, higher rates of recurrent thrombosis and thrombosis-related morbidity, and decreased quality of life.[4,5]

The pathogenesis of pediatric cancer-related thrombosis is multifactorial and may reflect genetic prothrombotic factors, tumor-related and treatment-related factors.[6] The role of hereditary thrombophilia in the development of thrombosis in children with cancer is poorly investigated and still unclear.

This study was undertaken to determine the prevalence of Factor V Leiden, prothrombin G20210A mutation and methylenetetrahydrofolate reductase (MTHFR) C677T mutation in children with hematological malignancies and malignant solid tumors, the frequency of cancer-associated thrombosis, and the role of inherited thrombophilic abnormalities in thrombotic events.

PATIENTS AND METHODS

Patients: Forty-seven children with primary cancer (36 with hematological malignancies and 11 with malignant solid tumors) referred to the Division of Hematology and Oncology, Department of Pediatrics, Clinical Hospital Centre Rijeka, Croatia, were included in the study. The following data were collected from medical records: gender, age at diagnosis, the type of cancer, previous and family history of thrombosis, insertion/type of a central venous line, and the presence/developmental time/site of thrombotic event. An informed written consent was obtained from the parents of all patients. The ethical approval from an institutional ethics board was obtained.

Methods: The samples were taken from peripheral blood in EDTA-containing tubes. The genomic DNA was prepared from whole blood with the NucleoSpin Blood kit (Macherey-Nagel GmbH & Co. KG, Düren, Germany). Genetic polymorphisms of Factor V G1691A (Factor V Leiden), Factor II-Prothrombin G20210A and MTHFR C677T were screened with real-time polymerase chain reaction (RT-PCR) on the Light Cycler® 1.5 Instrument, Roche Diagnostics, Germany. Tests were performed by binding specific DNA probes marked with fluorescent colors during PCR and melting curve analysis of marked PCR products, according to the manufacturer's instructions. Plasma homocysteine levels were not assessed routinely.

Statistical analysis: Descriptive statistics was used to summarize data. The results were compared with the relative frequencies of heterozygous and homozygous variants of each polymorphism in general population. Fisher's exact test was used to compare the prevalence of Factor II and Factor V Leiden polymorphism between boys and girls with cancer, and between children with hematological malignancies and solid tumors. The Chi-squared test was used to describe MTHFR genotype distribution in boys and girls with cancer, and between patients with hematological malignancies and solid tumors. P value of < 0.05 was considered statistically significant.

RESULTS

Thirteen (27.7%) girls and 34 (72.3%) boys were included in the study. The median age of patients was 8.8 years (range 0.4 – 19.3 years). Thirty-six patients had hematological malignancies (acute lymphoblastic leukemia [ALL] = 26, acute myeloid leukemia = 2, non-Hodgkin lymphoma = 7, Hodgkin lymphoma = 1) and 11 patients had solid tumors (malignant brain tumor = 3, soft tissue sarcoma = 3, osteosarcoma = 2, Ewing sarcoma = 1, neuroblastoma = 1, nasopharyngeal carcinoma = 1). All patients had inserted central venous lines: 33 patients had Broviac catheter and 14 had Port-a-cath.

Three (6.4%) patients (all boys) had heterozygous Factor II G20210A mutation, while no homozygosity was detected. Heterozygous Factor V Leiden was identified in 4 (8.5%) children (2 boys and 2 girls) with cancer, and no homozygous Factor V Leiden was found. MTHFR C677T heterozygosity was present in 21 (44.7%) patients, and homozygosity in 3 (6.4%). Six (46.2%) girls and 15 (44.1%) boys were heterozygous for MTHFR C677T, while 1 (7.7%) girl and 2 (5.9%) boys were homozygous. There was no statistical significance in the prevalence of FII G20210A mutation (Fisher's exact test, P=0.550), Factor V G1691A mutation (Fisher's exact test, P=0.304), and MTHFR C677T mutation (Chi-squared test, P=0.928) between male and female patients.

Two (5.6%) patients with hematological malignancies and one (9.1%) with solid tumor had heterozygosity for Factor II G20210A mutation, while no homozygosity was found. Factor V Leiden was identified in 3 (8.3%) children with hematological malignancies, and in 1 (9.1%) with solid tumor, and all patients had heterozygous form. Heterozygous MTHFR C667T mutation was identified in 14 (44.4%) children with hematological malignancies and in 5 (45.5%) children with solid tumors, while 2 (5.6%) patients with hematological malignancies and 1 (9.1%) with solid tumor had MTHFR C667T homozygosity. No statistical significance was found in the prevalence of Factor II G20210A mutation (Fisher's exact test, P=0.560), Factor V Leiden (Fisher's exact test, P=01.000) and MTHFR C667T mutation (Chi-squared test, P=0.936) between patients with hematological malignancies and solid tumors.

Combined thrombophilic defect were detected in 5 patients. Four children had heterozygous Factor V Leiden and MTHFR C667T heterozygosity. One patient had a combination of heterozygosity for Factor II G20210A and for MTHFR C667T heterozygosity. A previous or family history of thrombosis were negative in all patients.

Four (8.5%) children (all boys) had documented thrombotic event during the course of the disease: right axillar and brachial vein thrombosis in a patient with non-Hodgkin lymphoma, right brachial vein thrombosis in a patient with neuroblastoma, right subclavian, axillary and brachial vein thrombosis in a patient with nasopharyngeal carcinoma, and right atrial thrombosis in a patient with osteosarcoma. No patient had a recurrent thrombosis. Two patients had heterozygous Factor V Leiden (both combined with heterozygous but no homozygous MTHFR C677T mutation), one patient had heterozygous MTHFR C677T mutation, and one patient had no thrombophilia gene mutation detected.

The characteristics of pediatric oncology patients with thrombosis are shown in Table 1.

DISCUSSION

The frequency of a specific genetic polymorphism varies from population to population, reflecting ancient adaptation to specific environments. The most common inherited forms of thrombophilia in the United States and European general population are heterozygosity for Factor V Leiden and heterozigosity for prothrombin G20210A mutation, with the reported prevalence approximately 3 to 8%, and 1 to 6% respectively.[7,8]Homozygosity for MTHFR is common worldwide with estimated prevalence 10 to 25% among Caucasians, and combinations of other thrombophilic risk factors with MTHFR homozygosity are not unusual.[9] Two case-control studies investigated the frequency of inherited thrombophilia in Croatian population. Alfirevic et al. reported the frequency of heterozygous Factor V Leiden in 2.9% healthy individuals, heterozygous Factor II G20210A in 6%, heterozygous MTHFR C677T in 57%, and homozygous MTHFR T677T in 7% healthy controls. They found more frequent Factor V Leiden heterozygosity in patients with the thromboembolic disease (16%) compared to controls.[10]Coen and coworkers found the prevalence of 4% for both Factor V Leiden and PT20210A in healthy subjects. The prevalence of Factor V Leiden and prothrombin G20210A was higher in patients with venous thromboembolism, 21% and 8% respectively.[11] No homozygous Factor V Leiden or Factor II mutation was found in both Croatian studies.[10,11]

We found the prevalence of 8.5% for heterozygous Factor V Leiden, 6.4% for heterozygous Factor II G20210A mutation, 44.7% for heterozygous MTHFR C677T, and 6.4% for homozygous MTHFR C677T in our cohort. Several studies on the prevalence of inherited thrombophilic factors in children with malignancies obtained similar results. Akin and al. reported data for 135 Turkish patients with leukemia aged 1 to 15 years; 11 (8%) children were heterozygous for Factor V G1691A mutation, and 7 (5.1%) were heterozygous for prothrombin G20210A mutation.[12]Nowak-Göttle and coworkers conducted a large German prospective study of the thrombotic risk in 301 children with newly diagnosed ALL aged 6 months to 17 years, and found a single thrombophilic factor in 55 (18.2%). After exclusion of 12 patients, in the remaining 289 consecutively admitted children, 20 (6.9 %) showed MTHFR T677T genotype, 11 (3.8%) were carriers of Factor V G1691A mutation (10 heterozygous and 1 homozygous), 5 (1.7%) had heterozygous prothrombin G20210A variant, 4 (1.4%) showed protein C deficiency, 4 (1.4%) had protein S deficiency, 2 (0.7%) had antithrombin deficiency, and 9 (3.1%) patients had familiarly elevated lipoprotein concentrations. In addition, combined thrombophilia

was found in further 10 (3.5%) patients. The prevalence of all tested thrombophilic factors was within the prevalence reported for healthy white population.[13] Elhasid et al. found thrombophilic polymorphisms in 43% (13/27) children with ALL of Arab and Israeli origin older than 12 months of age. Prothrombin G20210A mutation was documented in 3 (11%), heterozygosity for Factor V Leiden in 5 (18.5%), and homozygosity for MTHFR C677T mutation in 5 (18.5%) children. One girl of Arab origin had triple thrombophilia. The higher prevalence was explained by higher frequency of heterozygosity for Factor V Leiden and Factor II mutation in the Israeli population of northern Israel.[14]

Studies on adult cancer patients showed conflicting results. Pihusch et al. found an increased prevalence of prothrombin G20210A mutation in patients with gastrointestinal carcinoma compared to normal population (5.7% versus 0.8%), while there was no difference in the prevalence of Factor V Leiden and MTHFR homozygosity, 6.9% and 9.7% respectively.[15] On the contrary, Turkish study reported significantly greater prevalence (30.2%) of Factor V Leiden in 43 cancer patients with thromboembolism compared to 3.7% those without, but no significant difference in the prevalence of prothrombin G20210A among the groups.[16] Battistelli and coworkers found no difference in the prevalence of Factor V Leiden and prothrombin G20210A gene mutation between 121 patients with gastric carcinoma and 130 healthy subjects from central Italy, matched for sex, age, and ethnicity (3.3% versus 4.6%, and 8.3% versus 6.1% respectively).[17] A large German population-based case-control study reported a 6-fold increased risk for colorectal cancer for homozygous carriers of Factor V Leiden compared with non-carriers. A reduced risk was demonstrated for heterozygous Factor V Leiden carriers and for heterozygous prothrombin G20210A carriers. No association was found between MTHFR polymorphisms with colon or rectal cancer.[18]Paspatis et al. reported a high frequency of activated protein C resistance (APCR) but no significant differences in the prevalence of Factor V Leiden or prothrombin G20210A mutation in Greek patients with colorectal cancer compared to colonoscopically selected controls.[19] Martinelli et al. studied the role of Factor V Leiden and prothrombin G20210A mutation for the development of thrombosis in 430 patients with hepatocellular carcinoma who underwent liver transplantation. They found 4-times higher risk of thrombosis in patients with inherited thrombophilia than in those without, and 6-times higher risk when the analysis was restricted to venous thrombosis. The presence of inherited thrombophilia in donors did not increase the risk of thrombosis of recipients.[20] Although dissimilarities in these studies could be partly explained by different ethnic background of subjects, they point the need of larger prospective multicentric investigations.

In our cohort, thrombosis was documented in 4 of 47 (8.5%) children with cancer, which is much higher than in the general pediatric population. The reported incidence of thrombosis ranges from 0.14 to 0.21 per 10,000 children per year, and 0.2 to 0.6% among hospitalized pediatric patients.[21] The majority of affected children have at least one underlying condition or trigger for thrombosis, the most common being central venous catheter, inherited thrombophilia, malignancy, congenital heart disease, chronic neuromuscular disease, surgery, major trauma, immobility, estrogen-containing contraceptives, obesity, and severe infection.[21-24]

The risk of thrombosis is considerably lower in children compared to adults. Several factors are considered to contribute to the small incidence of pediatric thrombosis: lower frequency of diseases causing vascular endothelium damage, less frequent exposure to acquired prothrombotic risk factors, and significant physiological differences in the coagulation system (lower plasma levels of vitamin K-dependent factors, higher levels of thrombin inhibitor and α2-macroglobulin, and reduced capacity to generate thrombin).[21,25]

The association between thrombosis and pediatric cancer is well established, and overall 25% of children with thrombosis have underlying diagnosis of cancer.[26] The reported prevalence of thrombosis in children with cancer ranges from 2 to 16%, while the occurrence of asymptomatic events is approximately 40%.[27-33] The risk is highest in children with ALL, followed by sarcoma and lymphoma, and the lowest risk is in children with brain tumors.[4,34,35] In the present study, the occurrence of thrombosis of 8.5% is in agreement with published data, but thrombosis was more frequent in children with solid tumors (3/11) compared to hematological malignancies (1/36).

The etiology of thrombosis in children with cancer is multifactorial, and includes patient-related predisposition (congenital thrombophilia), disease-related factors and treatment-related factors. Cancer may be

considered a hypercoagulable state. Tumor cells express tissue factor, procoagulant proteins, metalloproteases, and molecules that can induce direct and indirect activation of coagulation. Several additional mechanisms, such as inflammatory, immune, and angiogenic responses, are involved.[36,37] Major risk factors for thrombosis in children with hematological malignancies include presence of central venous catheter, older age, prothrombotic genetic defects, non-O blood group, obesity, and medications (asparaginase, concomitant use of steroids, anthracyclines).[4,38-40] Proposed thrombotic risk factors in children with solid tumors include presence of central venous line, age > 10 years, certain types and sites of tumor, metastatic disease, thrombophilia, obesity, and type of treatment (surgery, radiation, anthracyclines and platinum).[4,31,41] Central venous catheters are the most important risk factor.[42] Reported rates of symptomatic catheter-related thrombosis range from 2.6 to 36.7%, and rates of asymptomatic catheter-related thrombosis range from 5.9 to 43%.[3,43] The pathogenesis of catheter-related thrombosis is not well characterized, and it may involve endothelial damage and local activation of blood coagulation.[44]The most common site is upper venous system, and lower extremities for non-catheter-related thrombosis.[42,45] Central nervous system thrombosis is more common in children with ALL, with approximately half of patients having sinus venous thrombosis.[33,42] The incidence of cerebral sinus venous thrombosis in pediatric ALL patients varies from 1.4 to 10.5%.[46-49] Right atrial thrombosis is reported in 2% of patients with symptomatic thrombosis.[50]

In our cohort study, 3 of 4 children with thrombosis were adolescents aged > 15 years. All patients had implanted central venous access devices, and all thrombotic events occurred during chemotherapy. Three patients had an upper extremity thrombosis, and one had right atrial thrombosis. All patients were male. Two patients had documented thrombophilic gene mutations, both heterozygous Factor Leiden (combined with MTHFR C677T heterozygosity).

The contribution of inherited thrombophilia to the occurrence of thrombosis in cancer patients has been documented. The two most common genetic causes of thrombophilia identified to date are Factor V Leiden and prothrombin gene mutation.[51,52] Although MTHFR C677T heterozygosity is very frequent polymorphism, it is proven that this gene alteration increases the risk of thrombosis only when results in hyperhomocysteinemia.[53] A meta-analysis of 17 prospective studies comprising 1752 pediatric patients with ALL reported the overall thrombotic risk of 5.2%. Prothrombotic genetic defects were studied in 557 children. Thirty-one thrombotic events were observed in 113 children affected by at least one genetic alteration, pointing an approximately 8-fold increased thrombotic risk (relative risk [RR]:8.5; 95% CI: 4.4-17.4) in all patients with inherited thrombophilia.[40] Similar results were reported by Nowak-Göttle and coworkers, who documented venous thrombosis in 46.5% (27/58) children with ALL carrying a prothrombotic defect compared to 2.2% (5/131) children with no identified prothrombotic defect ($P < 0.0001$; chi-square 137.0). Homozygous MTHFR mutation with hyperhomocysteinemia was diagnosed in 12.5% (4/32) children with thrombosis, and in further 9.4% (3/32) patients combined with Factor V Leiden or increased lipoprotein A concentrations. In addition, an increased risk of thrombotic complications was clearly demonstrated in leukemia patients with combined prothrombotic risk factors compared to patients with single alterations.[13] The study of Knöfler et al. included 77 children with malignancies; in 11 (14%) of them catheter-related thrombosis was detected. Prothrombotic genetic defects were found in 23% (17/77) of all patients, and in 7 of 11 (64%) patients with thrombosis. Three children had combined defects (heterozygous Factor V G1691A mutation combined with heterozygous prothrombin G20210A variant, protein S deficiency or hyperlipoproteinemia), and 4 had a single defect (heterozygous Factor V G1691A mutation, heterozygous prothrombin G20210A mutation, hyperlipoproteinemia, and protein C deficiency type I).[54] Ünal and coworkers evaluated inherited and acquired prothrombotic risk factors in 37 children with malignancies and thrombosis. Congenital defects were detected in 15 (40%) patients: 8 had heterozygous Factor V G1691A mutation, 1 had heterozygous prothrombin G20210A mutation, 4 had lipoprotein(a) elevation, 1 had decreased protein S level, and 1 had decreased protein C level. The risk of thrombosis increased when accompanied by additional prothrombotic risk factors.[55] A large population-based study in Israel on 1191 children with ALL reported venous thromboembolism in 89 (7.5%) children. Thrombophilia screening was performed in 584 children, and findings were positive in 84 (14.4%). Patients with thrombophilia had significantly more thrombotic events compared to children without thrombophilia ($p < 0.001$).[56] Other studies failed to show any impact

of thrombophilic gene mutations on thrombosis risk in patients with cancer.[42,57-60] Thus, the impact of inherited thrombophilic markers on the development of thrombosis in pediatric oncology patients has not been completely clarified.

Our study confirms the higher occurrence of thrombosis in children with cancer. Although it has considerable limitations in terms of retrospective design, small number of patients, heterogenous diagnoses, and a limited panel of tested genetic prothrombotic traits, the results suggest that inherited thrombophilia could be implicated in increasing the risk of thrombosis in children with cancer. Larger multicenter prospective studies, development of guidelines for thrombophilia screening, identification of high-risk groups, individualized reevaluation of additional prothrombotic risk factors, and appropriate measures might help in the prevention and early intervention of thrombotic events.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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REFERENCES

1. Abdol Razak NB, Jones G, Bhandari M, Berndt MC, Metharom P. Cancer-Associated Thrombosis: An Overview of Mechanisms, Risk Factors, and Treatment. *Cancers (Basel)* . 2018;10(10):380.

2. Fernandes CJ, Morinaga LTK, Alves JL Jr, et al. Cancer-associated thrombosis: the when, how and why. *Eur Respir Rev.*2019;28(151):180119.

3. Ko RH, Thornburg CD. Venous Thromboembolism in Children with Cancer and Blood Disorders. *Front Pediatr* . 2017;5:12.

4. Athale UH, Yang JYK, Chan AKC. Thromboembolism in children with cancer. In: UpToDate, Pappo AS, O'Brien S (Eds), UpToDate, Waltham, MA. (Last updated on May 17, 2021; Accessed on February 02, 2022.)

5. Raffini L, Huang YS, Witmer C, Feudtner C. Dramatic increase in venous thromboembolism in children's hospitals in the United States from 2001 to 2007. *Pediatrics.* 2009;124(4):1001-1008.

6. Barg AA, Kenet G. Cancer-associated thrombosis in pediatric patients. *Thromb Res.* 2020;191 Suppl 1:S22-S25.

7. Kujovich JL. GeneReviews. Seattle (WA): University of Washington, Seattle; 2018. Factor V Leiden thrombophilia.

8. Kujovich JL. GeneReviews. Seattle (WA): University of Washington, Seattle; 2006. Prothrombin-related thrombophilia.

9. Rosenson RS, Smith CC, Bauer KA. Overview of homocysteine. In: UpToDate, Freeman MW (Ed), UpToDate, Waltham, MA. (Last updated on December 06, 2021; Accessed on February 21, 2022.)

10. Alfirevic Z, Simundic AM, Nikolac N, et al. of factor II G20210A, factor V Leiden, MTHFR C677T and PAI-1 5G/4G polymorphism in patients with venous thromboembolism: Croatian case-control study. *Bioch Med.* 2010;20(2):229-235.

11. Coen D, Zadro R, Honovic L, Banfic L, Stavljenic Rukavina A. Prevalence and association of the factor V Leiden and prothrombin G20210A in healthy subjects and patients with venous thromboembolism. *Croat Med J.* 2001;42(4):488-492.

12. Akin DF, Sipahi K, Kayaalp T, et al. Factor V Leiden and prothrombin 20210A mutations among Turkish pediatric leukemia patients. *Leuk Res Treatment.* 2012;2012:345-349.

13. Nowak-Göttl U, Wermes C, Junker R, et al. Prospective evaluation of the thrombotic risk in children with ALL carrying the MTHFR TT 677 genotype, the prothrombin G20210A variant, and further prothrombotic risk factors. *Blood.* 1999;93(5):1595-1599.

14. Elhasid R, Lanir N, Sharon R, et al. Prophylactic therapy with enoxaparin during L-asparaginase treatment in children with ALL. *Blood Coagul Fibrinolysis.* 2001;12(5):367-70.

15. Pihusch R, Danzl G, Scholz M, et al. Impact of thrombophilic gene mutations on thrombosis risk in patients with gastrointestinal carcinoma. *Cancer.* 2002;94(12):3120-3126.

16. Eroglu A, Ulu A, Cam R, Kurtman C, Akar N. Prevalence of Factor V 1691 G-A (Leiden) and prothrombin G20210A polymorphisms and the risk of venous thrombosis among cancer patients. *J Thromb Thrombolysis.*2007;23(1):31-34.

17. Battistelli S, Stefanoni M, Genovese A, Vittoria A, Cappelli R, Roviello F. Prevalence of factor V Leiden and prothrombin G20210A in patients with gastric cancer. *World J Gastroenterol.*2006;12(26):4179-4180.

18. Vossen CY, Hoffmeister M, Chang-Claude JC, Rosendaal FR, Brenner H. Clotting factor gene polymorphisms and colorectal cancer risk. *J Clin Oncol.* 2011;29(13):1722-1727.

19. Paspatis GA, Sfyridaki N, Papanikolaou N, et al. Resistance to activated protein C, Factor V Leiden and the prothrombin G20210A variant in patients with colorectal cancer. *Pathophysiol Haemost Thromb.*2002;32:2-7.

20. Martinelli I, Ponziani FR, Maino A, et al Thrombosis after liver transplantation for hepatocellular carcinoma. *PLoS One.*2017;12(10):e0186699.

21. Albisetti M, Chan AKC. Venous thrombosis and thromboembolism (VTE) in children: Risk factors, clinical manifestations, and diagnosis. In: UpToDate, O'Brien S (Ed), UpToDate, Waltham, MA. (Last updated on February 03, 2022; Accessed on February 12, 2022.)

22. Tuckuviene R, Christensen AL, Helgestad J, Johnsen SP, Kristensen SR. Pediatric venous and arterial noncerebral thromboembolism in Denmark: a nationwide population-based study. *J Pediatr.*2011;159(4):663-669.

23. Takemoto CM, Sohi S, Desai K, et al. Hospital-associated venous thromboembolism in children: incidence and clinical characteristics. *J Pediatr.* 2014;164(2):332-338.

24. Giordano P, Grassi M, Saracco P, et al; Paediatric Thrombosis Working Group of the RITI. Paediatric venous thromboembolism: a report from the Italian Registry of Thrombosis in Children (RITI). *Blood Transfus.* 2018;16(4):363-370.

25. Jinks S, Arana A. Venous thromboembolism in paediatrics. *BJA Educ.* 2019;19(9):305-312.

26. Athale UH, Chan AK. Thromboembolic complications in pediatric hematologic malignancies. *Semin Thromb Hemost.*2007;33(4):416-426.

27. Lipay NV, Zmitrovich AI, Aleinikova OV. Epidemiology of venous thromboembolism in children with malignant diseases: a single-center study of the Belarusian Center for Pediatric Oncology and Hematology.*Thromb Res.* 2011;128(2):130–134.

28. Athale U, Siciliano S, Thabane L, et al. Epidemiology and clinical risk factors predisposing to thromboembolism in children with cancer.*Pediatr Blood Cancer.* 2008;51(6):792-797.

29. Pelland-Marcotte MC, Pole JD, Kulkarni K, et al. Thromboembolism Incidence and Risk Factors in Children with Cancer: A Population-Based Cohort Study. *Thromb Haemost.* 2018;118(9):1646-1655.

30. Athale U, Cox S, Siciliano S, Chan AK. Thromboembolism in children with sarcoma. *Pediatr Blood Cancer.* 2007;49(2):171-176.

31. Schiavetti A, Foco M, Ingrosso A, Bonci E, Conti L, Matrunola M. Venous thrombosis in children with solid tumors. *J Pediatr Hematol Oncol.* 2008;30(2):148-152.

32. Paz-Priel I, Long L, Helman LJ, Mackall CL, Wayne AS. Thromboembolic events in children and young adults with pediatric sarcoma. *J Clin Oncol.* 2007;25(12):1519-1524.

33. Piovesan D, Attard C, Monagle P, Ignjatovic V. Epidemiology of venous thrombosis in children with cancer. *Thromb Haemost.*2014;111:1015–1021.

34. Athale UH, Nagel K, Khan AA, Chan AK. Thromboembolism in children with lymphoma. *Thromb Res.* 2008;122(4):459-465.

35. Tabori U, Beni-Adani L, Dvir R, et al. Risk of venous thromboembolism in pediatric patients with brain tumors. *Pediatr Blood Cancer.* 2004;43(6):633-636.

36. Castle J, Blower E, Kirwan CC. Update on the role of circulating tumour cells in cancer-associated thrombosis. *Thrombosis Update.*2021;5:100066.

37. Rubio-Jurado B, Sosa-Quintero LS, Guzmán-Silahua S, García-Luna E, Riebeling-Navarro C, Nava-Zavala AH. The prothrombotic state in cancer.*Adv Clin Chem.* 2021;105:213-242.

38. Spavor M, Halton J, Dietrich K, et al. Age at cancer diagnosis, non-O blood group and asparaginase therapy are independently associated with deep venous thrombosis in pediatric oncology patients: A risk model. *Thromb Res.* 2016;144:27-31.

39. Prasca S, Carmona R, Ji L, et al. Obesity and risk for venous thromboembolism from contemporary therapy for pediatric ALL.*Thromb Res.* 2018;165:44-50.

40. Caruso V, Iacoviello L, Di Castelnuovo A, et al. Thrombotic complications in childhood ALL: a meta-analysis of 17 prospective studies comprising 1752 pediatric patients. *Blood.*2006;108(7):2216-2222.

41. Athale U. Thrombosis in pediatric cancer: identifying the risk factors to improve care. *Semin Thromb Hemost.* 2007;33(4):416-426.

42. Albisetti M, Kellenberger CJ, Bergsträsser E, et al. Port-a-cath-related thrombosis and postthrombotic syndrome in pediatric oncology patients. *J Pediatr.* 2013;163(5):1340-1346.

43. Wiegering V, Schmid S, Andres O, et al. Thrombosis as a complication of central venous access in pediatric patients with malignancies: a 5-year single-center experience. *BMC Hematol.* 2014;14(1):18.

44. Dentali F, Gianni M, Agnelli G, Ageno W. Association between inherited thrombophilic abnormalities and central venous catheter thrombosis in patients with cancer: a meta-analysis. *J Thromb Haemost.* 2008;6(1):70-75.

45. Schoot RA, van de Wetering MD, et al; DCOG-Aristocaths Supportive Care Working Group. Prevalence of Symptomatic and Asymptomatic Thrombosis in Pediatric Oncology Patients With Tunneled Central Venous Catheters. *Pediatr Blood Cancer.* 2016;63(8):1438-1444.

46. Musgrave KM, van Delft FW, Avery PJ, et al. Cerebral sinovenous thrombosis in children and young adults with acute lymphoblastic leukaemia – a cohort study from the United Kingdom. *Br J Haematol.* 2017;179(4):667-669.

47. Ranta S, Tuckuviene R, Mäkipernaa A, et al. Cerebral sinus venous thromboses in children with acute lymphoblastic leukaemia - a multicentre study from the Nordic Society of Paediatric Haematology and Oncology. *Br J Haematol.* 2015;168(4):547-552.

48. Ghanem KM, Dhayni RM, Al-Aridi C, et al. Cerebral sinus venous thrombosis during childhood ALL therapy: Risk factors and management.*Pediatr Blood Cancer.* 2017;64(12):e26694.

49. El-Khoury H, Saifi O, Haddad S, et al. Treatment-induced cerebral sinus venous thrombosis in childhood acute lymphoblastic malignancies: New risk factors to consider. *Pediatr Blood Cancer.*2021;68(11):e29210.

50. Athale UH, Chan AK. Thrombosis in children with ALL: part I. Epidemiology of thrombosis in children with ALL. *Thromb Res.*2003;111(3):125-131.

51. Decousus H, Moulin N, Quenet S, et al. Thrombophilia and risk of venous thrombosis in patients with cancer. *Thromb Res.* 2007;120 Suppl 2:S51-61.

52. Horowitz N, Brenner B. Thrombophilia and cancer. *Pathophysiol Haemost Thromb.* 2008;36(3-4):131-136.

53. Khan S, Dickerman JD. Hereditary thrombophilia. *Thromb J.*2006;4:15.

54. Knöfler R, Siegert E, Lauterbach I, et al. Clinical importance of prothrombotic risk factors in pediatric patients with malignancy–impact of central venous lines. *Eur J Pediatr.*1999;158 Suppl 3:S147-150.

55. Ünal S, Varan A, Yalçin B, Büyükpamukçu M, Gürgey A. Evaluation of thrombotic children with malignancy. *Ann Hematol.*2005;84(6):395-399.

56. Barzilai-Birenboim S, Nirel R, Arad-Cohen N, et al. Venous Thromboembolism and Its Risk Factors in Children with ALL in Israel: A Population-Based Study. *Cancers (Basel).* 2020;12(10):2759.

57. Sifontes MT, Nuss R, Hunger SP, Wilimas J, Jacobson LJ, Manco-Johnson MJ. The factor V Leiden mutation in children with cancer and thrombosis. *Br J Haematol* . 1997;96(3):484-489.

58. Ruud E, Holmstrøm H, Natvig S, Wesenberg F. Prevalence of thrombophilia and central venous catheter-associated neck vein thrombosis in 41 children with cancer–a prospective study. *Med Pediatr Oncol.* 2002;38(6):405-410.

59. Mauz-Korholz C, Junker R, Gobel U, Nowak-Gottl U. Prothrombotic risk factors in children with ALL treated with delayed E. coli asparaginase (COALL-92 and 97 protocols). *Thromb Haemost.* 2000;83(6):840-843.

60. Mitchell LG, Andrew M, Hanna K, et al; Prophylactic Antithrombin Replacement in Kids with ALL Treated with Asparaginase Group (PARKAA). A prospective cohort study determining the prevalence of thrombotic events in children with ALL and a central venous line who are treated with L-asparaginase: results of the Prophylactic Antithrombin Replacement in Kids with ALL Treated with Asparaginase (PARKAA) Study. *Cancer.*2003;97(2):508-516.

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