UDC 575
DOI: 10.2298/GENSR1102371D
Original scientific paper

FV LEIDEN, FII G20210A AND MTHFR C677T MUTATIONS IN PATIENTS WITH LOWER OR UPPER LIMB DEEP VEIN THROMBOSIS

Valentina DJORDJEVIC[1], Iva PRUNER[1], Ljiljana RAKICEVIC[1], Mirjana KOVAC[2,3], Danijela MIKOVIC[2], Predrag MILJIC[2,4], Nebojsa ANTONIJEVIC[2,5], and Dragica RADOJKOVIC[1]

[1]Institute of Molecular Genetics and Genetic Engineering, University of Belgrade
[2]School of Medicine, University of Belgrade, Serbia
[3]Blood Transfusion Institute of Serbia, Haemostasis Department, Belgrade, Serbia
[4]Clinic of Hematology, Clinical Center of Serbia, Belgrade, Serbia
[5]Clinic for Cardiology, Clinical Centre of Serbia, Belgrade, Serbia

Djordjevic V., I. Pruner, Lj. Rakicevic, M. Kovac, D.Mikovic, P. Miljic, N.Antonijevic, and D. Radojkovic (2011): *FV leiden, FII G20210A and MTHFR C677T mutations in patients with lower or upper limb deep vein thrombosis* - Genetika, Vol 43, No. 2, 371 -380.

Deep vein thrombosis (DVT) is a multifactorial disease that occurs with frequency of 1/1000 per year. The FV Leiden, FII G20210A and MTHFR C677T mutations represent genetic factors for the occurrence of vein thrombosis. The goal of this study was to determine the frequency of these mutations in patients with DVT of upper and lower limbs. The study encompassed 119 patients divided in two groups. The group of patients with

Corresponding author: Iva Pruner,Laboratory for Molecular Biology,Institute of Molecular Genetics and Genetic Engineering, Vojvode Stepe 444a,P.O. Box 23,11010 Belgrade,Serbia,phone: + 381 11 3976658,fax: + 381 11 3975808 e-mail: iva.pruner@gmail.com

the lower limbs thrombosis included 77 patients, while the upper limbs thrombosis group included 42 patients. The presence of FV Leiden, FII G20210A and MTHFR C677T mutations was detected by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) analysis.

In patients with DVT of lower limbs, the frequency of FV Leiden mutation was 26,0% in heterozygous form and 1,3% in homozygous form. In the group of patients with DVT of upper limbs, the frequency of heterozygous carriers was 7.2%. In patients with DVT of lower limbs, FII G20210A mutation occurred in heterozygous form in 15.6% subjects, and in the group with DVT of upper limbs the frequency was 7.2% in heterozygous and 2.3% in homozygous form. The frequency of MTHFR C677T mutation in patients with lower limbs DVT was 42.8% in heterozygous form and 13% in homozygous form, while in the group of patients with upper limbs DVT, the frequency was 52.4% in heterozygous form and 9.5% in homozygous form.

The FV Leiden and FII G20210A mutations represent significant risk factors for the occurrence of DVT of lower limbs. These mutations are less frequent in DVT of upper limbs and more extensive further studies are needed to determine their potential role. The MTHFR C677T mutation represents less significant risk factor for lower limb DVT and should be taken into account only in cases when it occurs in combination with other risk factors.

Key words: FV Leiden, FII G20210A, MTHFR C677T, risk factors, vein thrombosis, upper and lower limb

INTRODUCTION

Deep venous thrombosis (DVT) is a common disease with an annual incidence of 1 in 1000 individuals and represents leading cause of death in pregnancy, postpartum period and in surgical patients. The mechanisms of DVT formation are not completely elucidated, considering the fact that they are caused by joint effect of enviromental and genetic risk factors (BICK, 2006).

Deep vein thrombosis (DVT) is a multifactorial disease which is predisposed by large number of acquired and genetic risk factors. Most common acquired risk factors are: pregnancy, surgery, trauma, long-term immobilization, immunization and malignancy (BICK, 2006). Since 1965, when the existence of familiar thrombosis has been shown, genetic risk factors have been extensively studied (EGEBERG 1965).

A single point mutation (G→A) at nucleotide position 1691 in the factor V gene results in a mutated form of factor V is known as factor V Leiden (CHUNILAL and BATES, 2009). This mutation results in a replacement of arginine residue 506 with a glutamine at one of the factor V cleavage sites for activated protein C (APC). Mutated factor V is resistant to inactivation by APC (BERTINA *et al.* 1994), resulting in increased thrombin generation and a hypercoagulable state. The prevalence of FV Leiden mutation in general population varies by 0% to 15% according to ethnicity

(REES *et al.* 1995). The risk of venous thrombosis is increased by 3 to 8 fold if the heterozygous form of factor V Leiden mutation is present and 80 fold if factor V Leiden mutation is present in homozygous form (REES *et al.* 1995).

A common genetic variation (a guanine to adenine transition at position 20210) in the 3'-untranslated region of the prothrombin gene has been associated with elevated prothrombin blood levels and increased incidence of venous thrombosis (ZIVELIN *et al.* 1997). It is present in approximately 18% of patients with personal and familiar history of DVT and ~3% of healthy Caucasian population (GADELHA *et al.* 2010, ROSENDAAL *et al.* 1998). The presence of FII G20210A mutation is associated with 3 fold increased risk of deep venous thrombosis (POORT *et al.* 1996)

The mutation in the gene coding for 5, 10-methylenetetrahydrofolate reductase (MTHFR) is a cause of moderate hyperhomocysteinemia which has high prevalence in individuals with venous thrombosis. The genetic defect is a cytosine to thymine missense mutation at nucleotide 677, which substitutes a valine for an alanine residue (CHUNILAL and BATES 2009). The mutation in the heterozygous or homozygous state correlates with reduced enzyme activity and increased thermolability (FROSST *et al.* 1995). It is highly prevalent in general population, with 30-50% in heterozygous form and 5-20% in homozygous form (FROSST *et al.* 1995).

The aim of this study was to determine the prevalence of FV Leiden, FII G20210A and MTHFR C677T mutations in patients with upper and lower limb deep venous thrombosis. As this is the first study of this kind in Serbian population, the additional goal was to evaluate the importance of these mutations as risk factors for development of upper or lower limb deep venous thrombosis.

MATERIALS AND METHODS

Our study included 119 patients (15-82 years, median 39 years). Patients were divided in two groups, depending on the localization of the thrombotic disorder. The group of patients with lower limb DVT consisted of 77 individuals (41 males and 36 females, 15-82 years, median 40 years), while the group of patient with upper limb DVT included 42 individuals (15 males and 27 females, 19-73 years, median 37 years). The study was approved by the hospital ethics committee.

Peripheral blood was taken on 3.8% Na-citrate as anticoagulant. Mutation detection was performed from the whole blood sample or purified genomic DNA. Genomic DNA was purified from 200μL of whole blood using QIAamp DNA blood mini kit (QIAGEN, Germany) according to manufacturer's protocol.

The FV Leiden and FII G20210A mutations were detected by multiplex polymerase chain reaction (PCR) on whole blood or isolated DNA, followed by simultaneous digestion with specific restriction enzymes *MnlI* and *HindIII* (New England BioLabs) (ĐORĐEVIĆ *et al.* 2001). The fragment of the MTHFR gene that included nucleotide 677 was amplified by PCR using previously described primers (FROSST *et al.* 1995), followed by digestion with *HinfI* (New England BioLabs) endonuclease. Normal and mutated alleles were distinguished by the size of the

restriction fragments, using electrophoresis on 10% polyacrylamide gels and visualised by silver staining (RADOJKOVIC and KUSIC 2000).

Statistical analyses were carried out by using Statistical Package for Social Sciences 13.0 for Windows (SPSS Inc., Chicago, Illinois, USA). The prevalence of each mutation was compared between patients and controls with the use of Fisher's exact. Data for control group from Serbian population, which consisted of 120 healthy blood donors (84 males and 36 females), was previously published (DJORDJEVIC *et al.* 2004). The odds ratio (OR) and 95% CI (*Confidence Interval*) were estimated separately for each mutation. Value $p \leq 0.05$ was considered statistically significant.

RESULTS

The results of our study are shown in Table 1. Among the 77 patients with lower limb DVT, 20 patients (26%) were detected with FV Leiden mutation in heterozygous form and one patient (1.3%) was homozygous for this mutation. There were 12 carriers (15.6%) of FII G20210A mutation in heterozygous state. No homozygous carriers were detected. Thirty-three patients (42.8%) were heterozygous and 10 patients (13.3%) were homozygous for MTHFR C677T mutation. Seventeen patients were carriers of two mutations either in homozygous or heterozygous form, while one patient had all three analyzed mutation in heterozygous form. There is a statistically significant frequency increase of heterozygous carriers of FV Leiden and FII G20210A mutations in patients with lower limb ($p=0.0007$ and $p=0.02$, respectively). Carriers of FV Leiden mutation have 5.6-fold increased risk for development on DVT (95%CI 2.26-14.18) compared to healthy population, while carriers of FII G20210A have 4.25-fold risk increase (95%CI 1.43-12.59). The MTHFR C677T mutation showed increased frequency, but this increase was no statistical significance compared with controls. Patients heterozygous and homozygous for MTHFR C677T mutation have slightly increased risk for DVT development (by 1.16 and 1.13 fold, respectively).

In the 42 patients with upper limb DVT, 3 heterozygous carriers (7.2%) of FV Leiden were detected. Three patients (7.2%) carried FII G20210A mutation in heterozygous and one (2.3%) in homozygous form. MTHFR C677T mutation was detected in 22 patients (52.4%) in heterozygous form and 4 patients (9.5%) in homozygous form. Five patients carried two mutations in heterozygous and/or homozygous form. There is no statistically significant increase of analyzed mutations in the group of patients with upper limb DVT compared to controls (DJORDJEVIC *et al.* 2004). Carriers of FV Leiden exhibit 1.24-fold greater risk for DVT occurrence (95%CI 0.31-5.04), while carriers of FII G20210A and MTHFR C677T exhibit 1.77 and 1.71 fold increased risk, respectively (Table 1).

Comparative statistical analysis of mutation frequencies in patients with DVT of lower limb versus patients with DVT of upper limb determined that FV Leiden mutation represents 4.56 fold greater risk (95%CI 1.27-16.41) for development of lower limb DVT in homozygous form and 1.67 fold (95%CI 0,067-41,82) in heterozygous form. Carriers of FII G202010A mutation are 2.4 fold likely

to develop lower limb DVT rather than upper limb DVT (95%CI 0.64-9.04). Also, homozygous carriers of MTHFR C677T mutations would rather develop lower limb DVT than upper limb DVT (1.42-fold, 95% CI 0.42-4.83).

Table 1. Genotype frequencies of FV Leiden, FII G20210A and MTHFR C677T mutations in patients with upper and lower limb deep vein thrombosis

	FV Leiden		**FII G20210A**		**MTHFR C677T**	
	heterozygous	homozygous	heterozygous	homozygous	heterozygous	homozygous
LL DVT (n=77)	20 (26.0%)	1 (1.3%)	12 (15.6%)	/	33 (42.8%)	10 (13.0%)
UL DVT (n=42)	3 (7.2%)	/	3 (7.2%)	1 (2.3%)	22 (52.4%)	4 (9.5%)
Control group (n=120)	7 (5.8%)	/	5 (4.2%)	/	47 (39.2%)	14 (11.7%)
P+	0.0007	0.39	0.02	/	0.79	0.82
OR+	5.66	4.72	4.25	/	1.16	1.13
95% CI+	2.26-14.18	0.19-117.49	1.43-12.59	/	0.65-2.08	0.47-2.69
P*	0.72	/	0.43	0.26	0.15	1.00
OR*	1.24	/	1.77	0.93	1.71	0.80
95% CI*	0.31-5.04	/	0.40-7.75	0.04-23.45	0.84-3.47	0.25-2.57
P†	0.05	1.00	0.38	0.36	0.61	0.77
OR†	4.56	1.67	2.4	0.17	0.68	1.42
95% CI†	1.27-16.41	0.067-41.82	0.64-9.04	0.007-4.48	0.32-1.45	0.42-4.83

DVT-deep vein thrombosis; LL-lower limb; UL-upper limb; OR- *Odds Ratio*; CI- *Confidence Interval*, P-probability

+: LL DVT compared to controls

*: UL DVT compared to controls

†: LL DVT compared to UL DVT

DISCUSSION

In this study we examined the frequency of FV Leiden, FII G20210A and MTHFR C677T mutations in patients with DVT of upper and lower limb in Serbian population. We noted the increased frequency of FV Leiden, FII G20210A and MTHFR C677T mutations in study groups, which implies that these mutations could be potential risk factors for the occurrence of DVT.

All three examined mutations are more frequent in the group of patients with DVT of lower limb in relation to our healthy population. Statistically significant increase is present only in the case of FV Leiden and FII G2010A mutations, which is in accordance with published studies. The Almawi *et al.* study (ALMAWI *et al.* 2005) that included 198 patients has shown that the presence of FV Leiden and FII G20210A mutations increases the risk of DVT 4 to 7 fold, whereas the MTHFR C677T mutation does not represent a significant risk factor *per se*, except when it occurs combined with FV Leiden or FII G20201A mutations (ALMAWI 2005 *et al.*,

KOVAC *et al.* 2008, KOVAC *et al.* 2010). On the other hand, the Kreidy and Irani-Hakime study, performed on the group of 162 patients with diagnosed DVT of lower limb (KREIDY and IRANI-HAKIME, 2009) shows that the acquired risk factors like operations, age, increased body weight, hormone therapies and pregnancy are much more significant for the occurrence of disease. Only 25 patients in this study had the hereditary thrombophilia, of which 16 carried FV Leiden mutation, 8 carried the MTHFR C677T mutation while one was the FII G20210A mutation carrier. Although it was shown that the increased level of homocysteine in blood poses a serious threat for the occurrence of DVT (ELDIBANY and CAPRINI 2007), such correlation was not observed in the case of MTHFR C677T mutation. In the MEGA study (BEZEMER *et al.* 2007) that encompassed nearly 5000 patients, it has been shown that there is no link between MTFHR C677T mutation and the occurrence of DVT, unless there is also a lowered level of vitamin B.

DVT of upper limb accounts for about 14% of thrombotic disorders (SPENCER *et al.* 2007). It was described in 19th century by Paget and von Schroetter, who presumed that traumatic fissures in the intima, which develop as the consequence of physical strain, lead to the consecutive activation of coagulation cascade (ZELL *et al.* 2001). For the last several decades, due to the use of central venous catheter, bone marrow transplantation, dialysis and parenteral feeding which represent greatest risk factors, the frequency of DVT of upper limb has increased (JOFFE *et al.* 2002). Other additional risk factors include: malignity, the use of oral contraceptives and thrombophilia (BLOM *et al.* 2005). Unlike the DVT of lower limb, literature data about DVT of upper limb are scarce, whilst the link between the thrombotic disorder and the DVT of upper limb has not been completely elucidated.

Our study has shown that increased frequency of FV Leiden, FII G20210A and MTHFR C677T mutations in the patients with the DVT of upper limb, but with no statistically significant differences in relation to control group. According to literary data, the frequency of coagulational abnormalities in the upper limb DVT varies from 8-61% (HENDLER *et al.* 2004). In most of the studies, FV Leiden is emanated as a significant risk factor, while the data concerning FII G20210A and MTHFR C677T mutations is incomplete or contradictory. MARTINELLI *et al.* as well as FLINTERMAN *et al*. observed that FV Leiden and FII G20210A mutations increase the risk of DVT of upper limb 3-6 fold, while the hyperhomocysteinemia and the use of oral contraceptives are not related to the upper limb DVT(MARTINELLI *et al.* 2004, FLINTERMAN *et al.* 2008). On the other hand, VAYA *et al.* (VAYA *et al.* 2003) found FII G2010A mutation to be a significant risk factor for DVT of upper limb in women who use oral contraceptives. These contradictory results are most probably due to the small sample size, non-equal distribution of patients' age, ethnic affiliation and different geographical prevalence of these mutations, as well as the presence of other acquired causes of DVT.

The comparative analysis of FV Leiden, FII G20210A and MTHFR C677T mutations frequency in the group of patients with DVT in upper and lower limb have shown that these mutations are a significant risk factors for the occurence of DVT of lower limb. One of the studies (LECHNER *et al.* 2008) has shown that the proportion

of FV Leiden carriers is significantly lower among those affected by DVT of upper limb compared to those affected by DVT of lower limb, while there was no significant difference in relation to the mutation in the prothrombin and MTHFR gene, which is in accordance with our results. Different frequency of these mutations in patients with DVT of both lower and upper limb implies the possibility of different pathophysiological mechanisms causing these two conditions. It has been reported that the patients with DVT of upper limb are younger and leaner compared to patients with DVT of legs. This observation is accordant with the findings that stress, age and body weigh are significant risk factors of DVT of lower limb (VAYA *et al.* 2003).

CONCLUSION

The FV Leiden and FII G20210A mutations represent significant risk factors for the occurrence of DVT of lower limb, while MTHFR C667T mutation is not a significant risk factor, but should be considered as an additional factor in case of occurrence along with other mutations and other risk factors for DVT. In cases of familiar thrombosis it is advised to perform routine testing for these mutations.

Based on our results, none of the examined mutations represent a statistically significant risk factor for the occurrence of upper limb DVT, but due to the increased frequency of these mutations in the group of patients and the contradictory literature data, it is needed to conduct the study that will include larger cohort of patients.

ACKNOWLEDGEMENT

This work was supported by grant No 173008 from the Ministry of Science and Technical Development of Serbia.

Received, January 24th2011
Accepted, July 05h 2011

REFERENCES

ALMAWI ,WY, H. TAMIM, R. KREIDY, G. TIMSON, E. RAHAL, M. NABULSI, RR. FINAN, N. IRANI-HAKIME (2005): A case control study on the contribution of factor V-Leiden, prothrombin G20210A, and MTHFR C677T mutations to the genetic susceptibility of deep venous thrombosis. J Thromb Thrombolysis;*19*(3):189-96.

BERTINA, RM, BP. KOELEMAN, T. KOSTER, FR. ROSENDAAL, RJ. DIRVEN, H. DE RONDE, PA. VAN DER VELDEN, PH. REITSMA (1994): Mutation in blood coagulation factor V associated with resistance to activated protein C. Nature; *369*(6475):64-7.

BEZEMER, ID, CJ. DOGGEN, HL. VOS, FR. ROSENDAAL (2007): No association between the common MTHFR 677C->T polymorphism and venous thrombosis: results from the MEGA study. Arch Intern Med; *167*(5):497-501.

BICK ,RL. (2006) Hereditary and acquired thrombophilic disorders. Clin Appl Thromb Hemost; 12(2):125-35.

BLOM, JW, CJ. DOGGEN, S. OSANTO, FR. ROSENDAAL (2005): Old and new risk factors for upper extremity deep venous thrombosis. J Thromb Haemost; *3*: 2471–8.

CHUNILAL, SD, SM. BATES. (2009): Venous thromboembolism in pregnancy: diagnosis, management and prevention. Thromb Haemost; *101*(3):428-38.

DJORDJEVIC, V, LJ. RAKICEVIC, M. GAGIC, A. NIKOLIC, A. SAVIC (2001): Simultaneous detection of factor V Leiden and factor II G20210A variant by a multiplex PCR on whole blood. Balc J Med Gen; *4*(1, 2):15-7.

DJORDJEVIC,V, LJ. RAKICEVIC, D. MIKOVIC, M. KOVAC, P. MILJIC, D. RADOJKOVIC, A. SAVIC (2004): Prevalence of factor V Leiden, factor V Cambridge, factor II G20210A and methylenetetrahydrofolate reductase in healthy and thrombophilic Serbian population. Acta Haematol; *112*:227-9.

ĐORĐEVIĆ, V, LJ. RAKIĆEVIĆ, M. SPASIĆ, P. MILJIĆ, D. MIKOVIĆ, M. KOVAČ, D. RADOJKOVIĆ (2005): Mutacije FV Leiden, FII G20210A i MTHFR C677T kao faktori rizika za nastanak tromboze dubokih vena u toku trudnoće ili puerperijuma. Vojnosanit Pregl; *62*(3): 201-5.

ELDIBANY, MM, JA. CAPRINI (2007): Hyperhomocysteinemia and thrombosis: an overview. Arch Pathol Lab Med; *131*(6):872-84.

EGEBERG, O. (1965): Inherited antithrombin deficiency causing thrombophilia. Thromb Diath Haemorrh;5;13:516-30.

FLINTERMAN, LE, FJM. VAN DER MEER, FR. ROSENDAAL, CJM. DOGGEN (2008): Current perspective of venous thrombosis in the upper extremity. J Thromb Haemost; *6*: 1262–6.

FROSST, P, HJ. BLOM, R. MILOS, P. GOYETTE, CA. SHEPPARD, RG. MATTHEWS, GJ. BOERS, M. DEN HEIJER, LA. KLUIJTMANS, LP. VAN DEN HEUVEL, et al (1995): A candidate genetic risk factor for vascular disease: a common mutation in methylenetetrahydrofolate reductase. Nat Genet;*10*(1):111-3.

GADELHA, T, V ROLDÁN, R LECUMBERRI, J TRUJILLO-SANTOS, R DEL CAMPO, R POGGIO, M MONREAL; RIETE INVESTIGATORS (2010): Clinical characteristics of patients with factor V Leiden or prothrombin G20210A and a first episode of venous thromboembolism. Findings from the RIETE Registry. Thromb Res; *26*(4):283-6.

HENDLER, MF, SS. MESCHENGIESER, AN. BLANCO, MF. ALBERTO, MJ. SALVIÚ, L. GENNARI, MA. LAZZARI (2004): Primary upper-extremity deep vein thrombosis: high prevalence of thrombophilic defects. Am J Hematol; *76*(4):330-7.

JOFFE, HV, SZ. GOLDHABER (2002): Upper-extremity deep vein thrombosis. Circulation; *106*: 1874–80.

KOVAC, M, D. MIKOVIC, N. ANTONIJEVIC, L. RAKICEVIC, V. DJORDJEVIC, D. RADOJKOVIC, I. ELEZOVIC (2008): FV Leiden mutation and risk of recurrent venous thromboembolism in Serbian population. J Thromb Thrombolysis; *25*:284-287.

KOVAC, M, G. MITIC, Z. MIKOVIC, N. ANTONIJEVIC, V. DJORDJEVIC, D. MIKOVIC, V. MANDIC, LJ. RAKICEVIC, D. RADOJKOVIC (2010). Type and location of venous thromboembolism in carriers of factor V Leiden or prothrombin G20210A mutation *versus* patients with no mutation. Clin Appl Thr Haemost; *16*(1):66-70.

KREIDY, R, N. IRANI-HAKIME (2009): Is thrombophilia a major risk factor for deep vein thrombosis of the lower extremities among Lebanese patients?. Vasc Health Risk Manag; 5:627-33.

LECHNER, D, C. WIENER, A. WELTERMANN, L. EISCHER, S. EICHINGER, PA. KYRLE (2008): Comparison between idiopathic deep vein thrombosis of the upper and lower extremity regarding risk factors and recurrence. J Thromb Haemost; *6*: 1269–74.

MARTINELLI, I., T. BATTAGLIOLI, P. BUCCIARELLI, SM. PASSAMONTI, PM. MANNUCCI (2004): Risk factors and recurrence rate of primary deep vein thrombosis of the upper extremities. Circulation; *110*:566-70.

POORT, SR, FR. ROSENDAAL, PH. REITSMA, RM. BERTINA (1996): A common genetic variation in 3`-untranslated region if the prothrombin gene is associated with elevated plasma prothrombin levels and an increase in venous thrombosis. Blood; *88*(10): 3698-703.

RADOJKOVIC, D, J. KUSIC (2000): Silver staining of denaturing gradient gel electrophoresis gels. Clin Chem; *46*(6 Pt1): 883-4.

REES, DC, M. COX, JB. CLEGG (1995): World distribution of factor V Leiden. Lancet. *346*(8983):1133-34.

ROSENDAAL, FR, CJ. DOGGEN, A. ZIVELIN, VR. ARRUDA, M. AIACH, DS. SISCOVICK, A. HILLARP, HH. WATZKE, F. BERNARDI, AM. CUMMING, FE. PRESTON, PH. REITSMA (1998): Geographic distribution of the 20210 G to A prothrombin variant. Thromb Haemost;*79*(4):706-8.

SPENCER, FA, C. EMERY, D. LESSARD, RJ. GOLDBERG, for the Worcester Venous Thromboembolism Study (2007): Upper extremity deep vein thrombosis: a community-based perspective. Am J Med; *120*:678-84.

VAYÁ, A, Y. MIRA, J. MATEO, C. FALCO, P. VILLA, A. ESTELLES, D. CORELLA, J. FONTCUBERTA, J. AZNAR (2003): Prothrombin G20210A mutation and oral contraceptive use increase upper-extremity deep vein thrombotic risk. Thromb Haemost;*89*(3):452-7.

ZELL, L, W. KINDERMANN, F. MARSCHALL, P. SCHEFFLER, J. GROSS, A. BUCHTER (2001): Paget-Schroetter syndrome in sports activities--case study and literature review. Angiology;*52*(5):337-42.

ZIVELIN, A, JH. GRIFFIN, X. XU, I. PABINGER, M. SAMAMA, J. CONARD, B. BRENNER, A. ELDOR, U. SELIGSOHN (1997): A single genetic origin for a common Caucasian risk factor for venous thrombosis. Blood; *89*(2):397-402.

MUTACIJE FV LEIDEN, FII G20210A I MTHFR C677T KAO FAKTORI RIZIKA ZA NASTANAK TROMBOZE DUBOKIH VENA GORNJIH I DONJIH EKSTREMITETA

Valentina ĐORĐEVIĆ[1], Iva PRUNER[1], Ljiljana RAKIĆEVIĆ[1], Mirjana KOVAČ[2], Danijela MIKOVIĆ[2], Predrag MILJIĆ[3], Nebojša ANTONIJEVIĆ[4,5] i Dragica RADOJKOVIĆ[1]

[1] Institut za molekularnu genetiku i genetičko inženjerstvo, Univerzitet u Beogradu, Beograd
[2] Institut za transfuziju krvi, Centar za ispitivanje poremećaja hemostaze, Beograd
[3] Institu za hematologiju, Klinički centar Srbije, Beograd
[4] Institut za kardiovaskularne bolesti, Klinički centar Srbije, Beograd

I z v o d

Tromboze dubokih vena (TDV) su multifaktorijalno oboljenje koje se javlja sa učestalošću 1/1000 stanovnika godišnje. Mutacije FV Leiden, FII G20210A i MTHFR C677T predstavljaju genetske faktore za nastanak venskih tromboza. Cilj ove studije je da se utvrdi učestalost FV Leiden, FII G20210A i MTHFR C677T mutacija kod bolesnika sa TDV gornjih ili donjih ekstremiteta. Studija je obuhvatila 119 bolesnika podeljenih u dve grupe. Grupa bolesnika sa TDV donjih ekstremiteta brojila je 77, a grupa sa TDV gornjih ekstremiteta 42 bolesnika. Prisustvo FV Leiden, FII G20210A i MTHFR C677T mutacija dokazivano je umnožavanjem odgovarajućeg fragmenta gena pomoću lančane reakcije umnožavanja polimerazom i digestijom dobijenih fragmenata restrikcionim enzimima (PCR-RFLP).Učestalost FV Leiden mutacije, u grupi bolesnika sa TDV donjih ekstremiteta, iznosila je 26,0% u heterozigotnom obliku i 1,3% u homozigotnom obliku. U grupi bolesnika sa TDV gornjih ekstremiteta, učestalost heterozigotnih nosilaca iznosila je 7,2%. Mutacija FII G20210A bila je prisutna kod 15,6% ispitanika u grupi bolesnika sa TDV donjih ekstremiteta u heterozigotnom obliku, dok je u grupi sa TDV gornjih ekstremiteta ova mutacija bila zastupljena sa 7,2% u heterozigotnom i 2,3% u homozigotnom obliku. Učestalost MTHFR C677T mutacije kod bolesnika sa TDV donjih ekstremiteta iznosila je 42,8% u heterozigotnom obliku i 13% u homozigotnom obliku, a kod TDV gornjih ekstremiteta 52,4% u heterozigotnom obliku i 9,5% u homozigotnom obliku.Mutacije FV Leiden i FII G20210A predstavljaju značajne faktore rizika za nastanak TDV donjih ekstremiteta. Kod TDV gornjih ekstremiteta ove mutacije su manje zastupljene i potrebno je sprovesti dalja istraživanja koja obuhvataju veći broj bolesnika. MTHFR C677T mutacija je manje značajan faktor rizika i treba ga razmatrati samo u slučajevima kada se pojavljuje u kombinaciji sa drugim faktorima rizika.

Primljeno 24. I. 2011.
Odobreno 05. VII. 2011.