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CLINICAL RESEARCH

Preconception Screening for Gene Polymorphisms Associated with Thrombophilia and Hyperhomocysteinemia Risk in Healthy Young Women

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Abstract

The frequency characteristics of the gene polymorphisms (FVL G1691A, FII G20210A, MTHFR C677T, MTHFR A1298C, MTRR A66G) associated with thrombophilia, hyperhomocysteinemia risk and different perinatal or pregnancy complications were studied. This examination was conducted among 130 planned-pregnancy healthy young women aged between 19 and 29 years. A gene mutation analysis was performed using a real-time polymerase chain reaction (real-time PCR). Factor V Leiden (FVL G1691A) and prothrombin gene (FII G20210A) mutations were not identified in the women surveyed. The frequency of the occurrence of the heterozygous FVL 1691G/A genotype associated with the risk of thrombosis during pregnancy was very low in these women (0.8%). The frequency of the MTHFR (methylenetetrahydrofolate reductase) 1298C/C mutant genotype was 11.5%, MTHFR 677T/T – 5.4%, and MTRR (methionine synthase reductase) 66G/G – 31.5%. A combination of the MTHFR 677TT/1298CC and MTHFR 677TT/MTRR 66GG mutant genotypes, which significantly increased the risk of pregnancy loss and neural tube defects, were found to occur in 0.8% of the cases.

We concluded that selective thrombophilia screening (FVL G1691A and FII G20210A) based on prior personal and/or family history of venous thromboembolism was more cost-effective than a universal preconception screening in all planning pregnancy women. However, in order to decrease the risk of congenital anomalies and pregnancy complications associated with folate dependent homocysteine metabolism, preconception care should include folate supplementation.

Keywords: *gene polymorphism; factor V Leiden; prothrombin gene mutation; methylenetetrahydrofolate reductase; methionine synthase reductase; preconception care.*

Introduction

The FVL G1691A and FII G20210A gene mutations are the common types of inherited thrombophilias. A systematic review and meta-analysis of the prospective cohort studies suggest a link between the maternal FVL G1691A/FII G20210A and placenta-mediated pregnancy complications including pregnancy loss, small for gestational age newborns, pre-eclampsia and placental abruption [1]. Folic acid is essential for normal embryogenesis and the course of pregnancy. The MTHFR gene C677T polymorphism and the MTRR gene A66G polymorphism are associated with various types of pregnancy complications: neuraltube defects, congenital heart defects, early and late miscarriages, uteroplacental insufficiency, Down syndrome, placental abruption and others [4-12].

The aim of the study was to investigate the incidence of the FVL G1691A, FII G20210A, MTHFR C677T/A1298C and MTRR A66G gene polymorphisms in planned-pregnancy healthy young women.

Material and Methods

The study was approved by the Chita State Medical Academy Ethics Committee. A cross-sectional descriptive study was conducted among 130 healthy planned-pregnancy young women aged between 19 and 29 years that did not have a pregnancy, family or individual history of venous thromboembolism. The FVL G1691A, FII G20210A, MTHFR C677T, MTHFR A1298C

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and MTRR A66G gene mutation analysis was conducted by real-time PCR from the samples randomly selected from the healthy individuals. Allele and genotype frequencies were calculated by direct counting. Hardy-Weinberg equilibrium was evaluated using a chi-square test.

Results

The FVL G1691A and FII G20210A gene mutations were not identified in the women surveyed. The frequency of the normal homozygous FVL 1691G/G genotype was significantly higher than the heterozygous FVL 1691G/A genotype ((99.2% (129/130) and 0.8% (1/130), respectively; *P<0.0001*)). The 1691A and 1691G allele frequencies for the FVL G1691A mutation were 0.996 and 0.004, respectively. Among these women, 98.5% (128/130) were homozygous (20210G/G) carriers of the FII G20210A, while 1.5% (2/130, *P<0.0001*) of them were heterozygous (20210G/A) carriers of the G20210A. The 20210A and 20210G allele frequencies for the FII G20210A mutation were 0.992 and 0.008, respectively (Table 1).

Table 1.

Frequencies of the FVL G1691A and FII G20210A gene polymorphisms in planned-pregnancy healthy young women (n=130).

Gene polymorphism	Genotype			Allele frequency *(P)*	
FVL G1691A	GG	GA	AA	G	A
	129 (99.2%)	1 (0.8%)	0	0.996	0.004
FII G20210A	GG	GA	AA	G	A
	128 (98.5%)	2 (1.5%)	0	0.992	0.008

In total, 115 (88.5%) of the women possessed the normal MTHFR A1298C genotype. The frequencies of the normal homozygotes (1298A/A) and the heterozygotes (1298A/C) were 46.2% (60/130) and 42.3% (55/130), respectively. The mutant alleles with the MTHFR 1298C/C genotype were found significantly less frequently when compared with the MTHFR 1298A/A and the MTHFR 1298A/C genotypes – in 15 samples (11.5%, *P<0.001*). The 1298A and 1298C allele frequencies for the MTHFR A1298C mutation were 0,673 and 0.327, respectively.

In this study, 123 of the 130 healthy young women had the normal MTHFR C677T genotypes. The MTHFR 677C/C genotype was detected in 70 (53.8%) of these women. The MTHFR 677C/T genotype was observed in 53 (40.8%) cases. The 677T/T homozygotes for the MTHFR C677T mutation were found in 7 (5.4%) cases. The 677C and 677T allele frequencies for the MTHFR C677T mutation were 0.742 and 0.258, respectively.

The frequency of the normal homozygous MTRR 66A/A genotype was significantly less than heterozygous MTRR 66A/G genotype ((27.7% (36/130) and 40.8% (53/130), respectively; *P<0.05*)). The 66G/G homozygotes for the MTRR A66G mutation were identified in the 41(31.5%) women surveyed. The 677A and 677G allele frequencies for the MTRR A66G mutation were 0.481 and 0.519, respectively.

The combined MTHFR 677TT/1298CC and MTHFR 677TT/MTRR 66GG mutant genotypes, which significantly increased the risk of pregnancy loss and neural-tube defects [9,13], were identified in 2 women: MTHFR 677TT/A1298CC – in 1 woman (1/130; 0.8%), MTHFR 677TT/MTRR 66GG – in 1 woman (1/130; 0.8%).

Discussion

The presence of the mutations that promote thrombophilia in the genes responsible for the folate metabolism and for plasma coagulation is often associated with pregnancy failures and may be the underlying cause in some cases. Incidentally, the FVL G1691A and FII G20210A gene mutations are associated mainly with recurrent spontaneous abortions, small for gestational age newborns, pre-eclampsia, and placental abruption [1,4,15]. The heterozygous genotypes for the FVL G1691A and FII G20210A mutations are associated with the risk of thrombosis in pregnancy of well under 1% (0.2% and 0.5%, respectively) [16]. In our study, the frequency of the heterozygous FVL 1691G/A genotype associated with a risk of thrombosis in pregnancy was very low (0.8%) among planned-pregnancy healthy young women.

Many studies in the literature have discussed the subject of the role of the MTHFR and MTRR gene mutations as a risk factor for neural tube defects, congenital heart defects, recurrent spontaneous early abortions and late miscarriages, uteroplacental insufficiency, Down syndrome, placental abruption and others [4-12]. As shown in Table 2, the frequency of the MTRR 66G/G genotype in the women surveyed was higher than the normal MTRR 66A/A genotype (31.5% and 27.7%, respectively). The frequencies of the MTHFR 677T/T and MTHFR 1298C/C genotypes, which contain mutant alleles, were significantly less than frequencies of the normal homozygous MTHFR 677C/C and MTHFR 1298A/A genotypes (53.8% vs 5.4% and 46.2% vs 11.5%, respectively). The findings of the earlier studies have shown that the combined presence of the MTHFR 677TT/1298CC and MTHFR 677TT/MTRR 66GG genotypes highly increased the risk of pregnancy loss and neural tube defects [9,13]. The combined presence of the MTHFR C677T/A1298C polymorphisms with mutant alleles was detected in 1 woman (0.8%). The prevalence of the combined MTHFR 677TT/MTRR 66GG genotype was 0.8%, too.

Table 2.

Frequencies of the MTHFR A1298C/C677T and MTRR A66G gene polymorphisms in planned-pregnancy healthy young women (n=130).

Gene polymorphism	Genotype			Allele frequency *(P)*	
MTHFR A1298C	AA	AC	CC	A	C
	60 (46.2%)	55 (42.3%)	15 (11.5%)	0.673	0.327
MTHFR C677T	CC	CT	TT	C	T
	70 (53.8%)	53 (40.8%)	7 (5.4%)	0.742	0.258
MTRR A66G	AA	AG	GG	A	G
	36 (27.7%)	53 (40.8%)	41 (31.5%)	0.481	0.519

Conclusion

The results of our study showed the absence of the FVL G1691A and FII G20210A mutations in healthy planned-pregnancy young women between 19 and 29 years of age. The frequency of the heterozygous FVL 1691G/A genotype associated with the risk of thrombosis in pregnancy was very

low in these women. We concluded that selective thrombophilia screening (FVL G1691A and FII G20210A) based on prior personal and/or family history of venous thromboembolism was more cost-effective than a universal preconception screening in all planning pregnancy women. The high frequency of the MTRR and MTHFR gene polymorphisms was observed in the healthy planned-pregnancy young women (MTRR 66G/G - 31.5%, MTHFR 1298C/C - 11.5%, and MTHFR 677T/T - 5.4%). However, in order to decrease the risk of congenital anomalies and pregnancy complications, preconception care should include folate supplementation.

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