Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Relevance of MTHFR gene variants with homocysteine level in Iraqi hypertensive Population

Salih Mahdi Al-Khafaji
Kufa University
Anwar Madlool Al-janabi
Kufa University
Dawood Ali Salim Dawood
alisalim@uowasit.edu.iq
Wasit University
Hamid Jaddoa Abbas
Al-Faiha'a Teaching Hospital

Research Article

Keywords: MTHFR gene, Hypertension, Homocysteine, Genotype, PCR-RFLP

Posted Date: May 29th, 2026

DOI: https://doi.org/10.21203/rs.3.rs-9684649/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License.
Read Full License

Additional Declarations: No competing interests reported.

Abstract

Background

Hypertension (HTN) is a multifactorial condition which is predetermined by genetic and environmental conditions. Variation differences in the methylenetetrahydrofolate reductase (MTHFR) gene could be a contributory factor to the high levels of homocysteine (Hcy) and high-risk for cardiovascular diseases. The objective was to investigate the relationship amongst MTHFR C677T and A1298C polymorphisms, homocysteine, and hypertension in a sample of Iraqi population.

Methods

A case control study was performed on 100 hypertensive patient and 150 normotensive controls. PCR-RFLP was applied in genotyping and ELISA to measure the plasma Hcy level. Genotype distribution and risk relationship had been evaluated using statistical analysis.

Results

The Hcy levels were in significant increase in hypertensive patients as compared to controls ($P < 0.001$). The polymorphism C677T was found to bring an elevated risk of HTN specifically to CT (OR = 2.1) and TT (OR = 8.09) genotypes. The A1298C was also strongly linked with A1298C (OR = 5.1) and AC (OR = 5.0) genotypes making individuals vulnerable. More common in patients were mutant alleles (T and C) that were associated with high levels of Hcy.

Conclusion

MTHFR polymorphisms have a close relationship with hypertension and hyperhomocysteinemia in the Iraqi population, and thus may be applicable in the risk assessment and patients' treatment.

Introduction

Hypertension (HTN), or high blood pressure, is defined as having systolic blood pressure (SBP) ≥ 140 mmHg or diastolic blood pressure (DBP) ≥ 90 mmHg. It is considered a risk factor leading to cardiovascular disease and associated with increased mortality from 6.8 million in 1990 to 10.8 million in 2019; recently, it was estimated to affect 33% of adults aged 30–79 worldwide [1]. In Iraq, HTN is a significant health problem with a prevalence rate of 35.6%; environmental and genetic predispositions are thought to contribute to the development and progression of HTN [2]. Hyperhomocysteinemia (HHcy) is one of the risk factors for atherosclerosis by which homocysteine (Hcy), is an intermediate substance formed during conversion of the essential amino acid methionine to cysteine by the action of methylenetetrahydrofolate reductase (MTHFR) enzyme. Hcy affects several biological and cellular

processes as cellular methylation and endothelial cells that located in the interior-most layer of blood vessels and forming vascular intima, which is essential for maintaining normal blood circulation and vascular homeostasis. In recent years, several studies have revealed that Hcy can damage the endothelium of blood vessels through different mechanisms that may promote atherosclerosis-related diseases. Many factors that influence the levels of Hcy in plasma comprise genetics, age, nutrition and disease states [3].

Genome wide association studies (GWAS) have recognized specific genes associated with blood pressure (BP) that could lead to personalized treatments for HTN depending on genetic characteristics. The earliest of such studies tested 2.5 million single nucleotide polymorphisms (SNPs) and identified eight genetic loci associated with BP, including a region near the gene encoding the folate-metabolizing enzyme MTHFR [4, 5]. Clinical studies have connected this gene with BP, meta-analyses of case control studies showing that the 677C→T and 1298A→C point mutations in MTHFR are associated with an increased risk of HTN by 36–87% [6–8].

Recent studies indicate that the MTHFR 677TT variant, present in approximately 10% of the global adult population, and associated with elevated Hcy concentrations. Increased Hcy is recognized as an independent risk factor for cardiovascular disease and may also contribute to the development of HTN [3, 9]. Hcy concentrations are regulated by interaction of both nutritional status and genetic factors [3]. Data has suggested that increased Hcy may also play a role in development of HTN, and may provide potential mechanism associated with vascular disease. Experimental facts has shown that elevated homocysteine has negative effects on the potent vasodilator [3], vascular smooth muscle, cell proliferation [10], impaired endothelial function [11], altered elasticity of the vascular wall and renal function [12], given that physiological factors such as peripheral resistance, arterial stiffness and impaired kidney function are also strong determinants of BP, it would therefore seem reasonable to predict an association between homocysteine and blood pressure.

In Iraq, up to now people with HTN is highly increased in numbers, evaluation of genetic predisposition among individuals is important for early diagnosis, risk stratification, and personalized treatment approaches.

The purpose of the study was to assess the importance of MTHFR gene mutations among patients with HTN in Iraq and determine whether the mutations influence the clinical outcome, such as BP and Hcy concentration.

Material and methods

Subject Groups

The present case–control study was conducted between March 2024 to January 2025 at molecular laboratory of Histology and Anatomy Department/ College of Medicine/ University of Kufa/ Najaf/ Iraq. The study was obtained the ethical approval committee of the College of Medicine / University of Kufa,

Iraq, under registration No. (MEC-191). The population study was involved 100 hypertensive patient were selected from internal medicine outpatient clinic at AL-Sadder Medical City teaching hospital, Kufa, Iraq, with an age range of 30 to 65 years. They were matched in age and sex with a control group of 150 normotensive individual with an age range of 29 to 67 year. All patients who participated in the study were diagnosed with HTN by physicians their BP exceeded 140 mmHg systolic and 90 mmHg diastolic as recorded on at least three occasions over two-month period prior to the administration of antihypertensive medication. Any subject suffered from diabetes mellitus, cardiac diseases, primary renal dysfunction, cancers and autoimmune diseases were excluded from the study. The NT individuals had no past or current history of HTN or cardiovascular disease and they had a BP of less than 140/90 mmHg. Personal consent was obtained from all participants before collecting blood samples.

Genotyping of MTHFR (C677T and A1298C) Gene Variant

From whole blood genomic was isolated by using DNA extraction kit ReliaPrep™ (Promega, Madison, WI, USA). The DNA was amplified by a DNA Thermal Cycler instrument (Biometra, Göttingen, Germany) polymerase chain reaction (PCR), using MTHFR (C677T) primers 5-TGAAGGAGAAG GTGTCTG CGGGA-3 for sense and 5-AGGACGGTGCGGTGAGAGTG-3 for antisense oligonucleotide. While MTHFR (A1298C) primers 5-TCCTCTTCCCCTGCCTTTG-3 for sense and 5-CCACTCCAGCATCACTCACTTT-3 for antisense oligonucleotide. The following singleplex PCR reaction used for each of C677T and A1298C: 1.75 mmol/L MgCl2, 0.2 mmol/L of deoxynucleotide triphosphates (dNTPs), 0.2 µmol/L forward and reverse primers, 0.16 g/L Bovin serum albumin (BSA), 1 unit Taq polymerase, 50 ng genomics DNA, made to a final volume of 25 µl with distilled water. The cycle steps were as follows: 1 cycle at 95°C (3 min) for an initial denaturation, followed by 35 cycles of denaturation (1min) at 94°C, primer annealing (1 min) at 65°C, primer extension (2 min) at 72°C and a final extension (10 min) at 72°C [13]. This amplification reactions resulted in the synthesis of fragment with 198 bp for C677T and 241 bp for A1298C.

MTHFR gene contains C > T substitutions at nucleotide 677, the variation recognized by HinfI (Promega, Madison, WI, USA) at 37°C for 4h. Accordingly, samples who were wild CC absent the mutation appear single band 198 bp, sample with heterozygous CT for the variation revealed both 198 bp and 175 bp bands, and homozygous TT sample revealed single band 175 bp band. On the other hand, MTHFR gene A to C substitutions at nucleotide 1298 created by MboII (Promega, Madison, WI, USA) at 37°C for 4 h, in MTHFR A1298C mutation, AA wild genotype appeared with only 241 bp, AC heterozygote genotype with 241 bp and 204 bp while CC homozygote mutant genotype with 204 bp.

RFLP-PCR technique was used to detect the variations among participants. The results of DNA fragments were visualized by UV transillumination after separating on 1% agarose gel stained by ethidium bromide as shown in Fig. 1 (A and B). Regarding to both mutants homozygotes (TT and CC) genotypes and heterozygotes (CT and AC) genotypes, the bands definitely disappeared on gel due to their small sizes (23 and 37 bp) respectively.

A plasma homocysteine levels were analyzed by using microplate enzyme immunoassay ELISA Biorad kit according to the manufacturer procedure, glycosylated hemoglobin (HbA1c) measured by using HbA1c fast test kit according to the manufacturer instructions, lipid profile: include serum total cholesterol, triglycerides, HDL- cholesterol, LDL- cholesterol, blood urea and serum creatinine measured with colorimetric methods by using commercial kits (BIOLABO SA, Maizy, France).

Statistical analysis

Statistical analysis was performed by using SPSS version 27.0, Armonk, New York, 2020. A continuous variables were expressed as a mean ± standard deviation (Mean ± SD). The students t-test and ANOVA were applied to compare variables means of continuous characteristics among genotypes. To assess the association between genotypes and allele frequencies of different inheritance models, the multinominal logistic regression was used in HTN and NT individuals. Categorical information of alleles and genotypes were expressed as frequency and odds ratio (OR) with 95% confidence intervals (C.I) were calculated. Hardy-Weinberg equilibrium was estimated for both groups. *P*-value < 0.05 was considered as statistically significant.

Results

General characteristics

A total of 250 participants were recruited in this study, comprising 100 patient and 150 healthy control. The statistical power was (1–β = 0.8086), which means the results are reliable. General characteristics of two groups were recorded; mean age of HTN were 48 ± 2.3 years and of NT were 47 ± 3.4 years. The HTN group had a significantly higher blood pressure (systolic BP = 160 ± 4.0 and diastolic BP = 95 ± 5.5) than NT group (systolic BP = 105 ± 3.0 and diastolic BP = 70 ± 5.0). Hcy levels were significantly higher among HTN when compared with NT group (*P* < 0.05). Other parameters including sex, age, BMI, lipid profile, glycosylated hemoglobin and renal functions tests were significantly not different between two groups (*P* > 0.05), this means that the groups were well-matched and that any possible confounding effects were reduced as shown in Table 1.

Table (1): General and clinical characteristics of hypertensive and normotensive groups

Variables	Patients	Control	*P* value
Sample size	100	150	--
Power (1-β err prob)	0.8086		
Sex: Male/Female	60/40	90 /60	0.07
Age at study (years)	48 ± 2.3	47 ± 3.4	0.11
BMI (kg/m^2)	26 ± 2.0	23 ± 1.7	0.06
B. P (mmHg)/ Systolic	160 ± 4.0	105 ± 3.0	< 0.001 ***
/ Diastolic	95 ± 5.5	70 ± 5.0	< 0.001 ***
Homocysteine (μM)	25.3 ± 2.1	9.0 ± 0.6	< 0.001 ***
Triglycerides (mg/dL)	128.3 ± 6.5	115.3 ± 9.8	0.13
Total cholesterol (mg/dL)	168.5 ± 6.4	149.8 ± 1.8	0.093
HDL cholesterol, mg/dL	40.3 ± 13.1	52.4 ± 7.3	0.08
LDL cholesterol, mg/dL	130.2 ± 7.4	94 ± 4.1	0.065
VLDL cholesterol, mg/dL	48.1 ± 4.4	46.9 ± 4.2	0.07
HbA1c (%)	5.1 ± 0.4	4.6 ± 0.9	0.1
Blood Urea(mg/dl)	40 ± 2.2	37 ± 3.6	0.19
Creatinine(mg/dl)	1.0 ± 0.1	0.7 ± 0.2	0.16

HT: Hypertensive; NT: Normotensive; BMI: Body Mass Index, B. P: Blood Pressure; HDL: High Density Lipoprotein; LDL: Low Density Lipoprotein; VLDL: Very Low-Density Lipoprotein and HbA1c: Glycosylated Hemoglobin.

Correlation of genotypes models of MTHFR (C677T and A1298C) gene mutations and HTN development

A significant observation after analyzing the MTHFR (C677T) polymorphism was reported. There was a notable correlation between hypertensive and normotensive groups as reflected in Table 2. The association of the CT genotype with increased risk of developing hypertension was observed with a 2-fold (OR 2.1; 95% CI: 1.2–3.7; P = 0.009) higher risk of developing hypertension compared to the control group. The frequency of TT genotype was significantly higher in patients with hypertension and had a significantly strong degree of association with the increased risk of hypertension (OR 8.09; 95% CI: 3.5–29.5, P = 0.001). In the dominant model (CT + TT vs CC), carriers of the T allele had a significantly increased risk of hypertension (OR 2.7; 95% CI: 1.6–4.7, *P* < 0.001), while a strong association was also shown in the recessive model (CC + CT Vs TT) also demonstrated a strong association (OR 7.2; 95% CI:

2.6–20, *P* < 0.001). Moreover, the prevalence of the T allele was greatly prevalent in hypertensive group indicating its role in the disease susceptibility.

Table (2): Risk of hypertension associated with MTHFR C677T genotype according to different models of inheritance

Genotype/Allele frequency of C677T SNP	Patients	Control	OR	(95% C.I)	*P* value
Co-dominant CC	35 (35%)	90 (60%)	1	Ref.	-
CT	45 (45%)	55 (36.7%)	2.1	1.2–3.7	0.009 **
TT	20 (20%)	5 (3.3%)	8.09	3.5–29.5	< 0.001 ***
Dominant CT + TT Vs CC	65 (65%)	60 (40%)	2.7	1.6–4.7	< 0.001 ***
Recessive CC + CT Vs TT	80 (80%)	145 (96.7%)	7.2	2.6–20	< 0.001 ***
C	115 (57.5%)	235 (78.3%)	1	Ref.	-
T	85 (42.5%)	65 (21.7%)	2.7	1.8–3.9	< 0.001 ***
H. W	0.427	0.326			

SNP, single nucleotide polymorphism; OR, odds ratio; CI, confidence interval; HWE, Hardy–Weinberg equilibrium.

P ≤ 0.05 considered significant.

On the same note, the polymorphism MTHFR A1298C was significantly linked to hypertension as shown in Table 3. The frequency of AC and CC genotypes was much greater in hypertensive population. Individuals with the AC genotype had a five-fold increased risk (OR 5.1; 95% CI: 2.8–9.4, *P* < 0.001), while those with the CC genotype exhibited a comparable risk (OR 5.0; 95% CI: 2.4–10.7, *P* = 0.0003). The dominant model (AC + CC vs AA) suggested a significantly increased risk (OR 6.7; 95% CI: 3.8–11.8, *P* < 0.001), while the recessive model (AA + AC vs CC) was also significant (OR 2.7; 95% CI: 1.4–5.6, *P* = 0.004). The frequency of the C allele was significantly increased in hypertensive patients, which adds to the evidence that the C allele is a genetic risk factor.

Table (3): Risk of hypertension associated with MTHFR A1298C genotype according to different models of inheritance

Genotype/Allele frequency of A1298C SNP	Patients	Control	OR	(95% C.I)	*P* value
Co-dominant	32 (32%)	104 (69.3%)	1	Ref.	-
AA					
AC	43 (43%)	30 (20%)	5.1	2.8–9.4	< 0.001 ***
CC	25 (25%)	16 (10.7%)	5.0	2.4–10.7	0.0003 **
Dominant AC + CC Vs AA	68 (68%)	46 (30.7%)	6.7	3.8–11.8	< 0.001 ***
Recessive AA + AC Vs CC	75 (75%)	134 (89.3%)	2.7	1.4–5.6	0.004 **
A	107 (107%)	238 (158.7%)	1	Ref.	-
C	93 (93%)	62 (41.3%)	5.3	3.5–8.2	< 0.001 ***
H. W	0.174	< 0.001			

SNP, single nucleotide polymorphism; OR, odds ratio; CI, confidence interval; HWE, Hardy–Weinberg equilibrium.

P ≤ 0.05 considered significant.

Association between MTHFR (C677T and A1298C) gene mutations and Hcy levels

Hcy serum concentration differed greatly based on genotype in both polymorphisms. In the case of the C677T variant, Hcy levels raised gradually in hypertensive patients in the CC, CT and TT genotypes, with the highest levels of Hcy in TT carriers (43.6 ± 3.7 μM). This tendency was observed in normotensive subjects but with lower concentrations. The A1298C polymorphism had Hcy levels that were highly expressed in all hypertensive patients, with the most levels representing the carriers of the CC genotypes (35.6 ± 4.3 μM). All the comparisons were extremely significant (P < 0.0001) which are indicated that MTHFR polymorphisms are strongly associated with high levels of Hcy Table 4.

Table (4): Comparison of serum Hcy levels (μM) between patients and control groups according to different models of inheritance.

SNP	Genotype	Patients	Control	*P* value
C677T	CC	18.8 ± 2.1	9.3 ± 1.1	< 0.0001 ***
	CT	36.2 ± 3.3	12.7 ± 1.7	< 0.0001 ***
	TT	43.6 ± 3.7	14.8 ± 2.3	< 0.0001 ***
A1298C	AA	17.4 ± 2.4	8.2 ± 2.6	< 0.0001 ***
	AC	28.3 ± 2.3	11.9 ± 1.6	< 0.0001 ***
	CC	35.6 ± 4.3	12.8 ± 1.5	< 0.0001 ***

SNP, single nucleotide polymorphism.

P ≤ 0.05 considered significant.

Discussion

HTN is an environmental and genetically determined multifactorial disease. It is a major risk factor for cardiovascular disease, with considerable morbidity and mortality. Furthermore, atherosclerosis is an independent predictor of both HTN-mediated organ damage and adverse cardiovascular outcomes [14–16]. Researchers have reported that genetic factors contribute to about 60% of HTN risks [5]. One of the enzymes is MTHFR, which plays a significant role in keeping the balance between the two amino acids, methionine and Hcy to avoid dysfunction of the cell due to the effect of Hcy, which increases risk for blood clots, heart disease and stroke [3]. The roles of the C677T and A1298C point mutations were observed to reduce the activity of MTHFR gene that encodes the MTHFR enzyme.

This study was conducted in non-relative subjects and the results revealed that there are no statistical differences in some of demographic characteristics between patients and healthy individuals in respect to age, sex, BMI, serum lipid profile, HbA1c, blood urea, and serum creatinine while other statistical analysis showed that there is a high significant difference in two groups regarding to BP and Hcy levels. These results agreed with Fowdar et al. in Australian population and Liu et al in Ningxia Hui nationality for Chinese population, who detected the association between Hcy and HTN earlier [17, 18]. Gai et al. had similar findings among the Chinese population [19].

Our results also showed that mutated homozygous TT genotypes of C677T and CC genotype of A1298C, so T and C mutant alleles were higher in patients compared to NT group in Co-dominant, dominant and recessive models of inheritance, these results consistent with similar studies of Arab population by Alghasham et al. who showed that T allele in CT and TT genotypes for C677T mutation and C allele in AC and CC genotypes for A1298C mutation were statistically significant in patients when compared with control group and Nassereddine et al. who discovered the homozygous mutant allele 677TT had a strong relationship with the risk of HTN. In addition, the other inheritance models depicted the same trend.[20, 21]. Also, meta-analysis of Hao et al. agree with our results, who found that MTHFR gene

mutation 677TT risk genotype in the overall population, Caucasians: Europeans, South Asians, Southeast Asians, East Asians, (allelic model, homozygote model, dominant model, and recessive model), and Africans [22]. Xu et al. also agree with our results who was found that blood pressure levels were significantly higher in participants with 677 TT genotype compared to those with CC or CT genotypes [23]. Indeed, homozygous mutation for the 677 C→T and 1298 A→ C in MTHFR gene causing decrease production of 5- methyltetrahydrofolate, the main methyl donor in alteration of Hcy to Met which lead to rise of Hcy in plasma [24]. 677 C→T, 1298A→C mutations is responsible for reduced MTHFR activity leading to accumulation of Hcy and it is found significantly in recessive homozygous genotypes [23].

Hcy metabolism is achieved by the enzyme MTHFR. This enzyme catalyzes the remethylation of Hcy into Met [19, 25]. Mutation on the gene coding MTHFR enzyme lead to decreasing the activity of the enzyme and augmented Hcy levels. Nowadays, the most common genetic cause of HHcy is known as the mutant allele 677C > T that leads to the reduction in the activity of MTHFR enzyme that has a significant role in the Hcy metabolism [19, 20]. These results consistent with our results that recorded high Hcy level in 677TT and 1298CC genotypes and the effect of C677T gene mutation on activity of an enzyme is greater than that of A1298C. Also, Xu et. al agree with our finding who had suggested that MTHFR C677T polymorphism could influence the level of Hcy and it may be strongly correlated with the prevalence of HTN [21]. Furthermore, Kuras et al., studied MTHFR C677T gene variant in Turkish population and concluded that this polymorphism is more common in hypertensive patients. The risk of HTN and Hcy levels may be increased by T allele frequencies and TT genotypes. Although A1298C, C allele frequency might increase the risk of HTN but CC genotype distributions and Hcy levels show no statistical significance on HTN [26]. According to Zhao et al., found in a cohort of hypertensive adults with an average 6-year follow-up, each 1 mol/L increase in Hcy was significantly associated with cardiovascular diseases mortality [27]. Regarding to homocysteine-lowering treatment, folic acid and vitamin B complex are thought to be effective for preventing atherosclerosis, cardiovascular diseases and neurodegenerative diseases [28].

As a result, it is unclear whether homocysteine is a biomarker or a reversible causal risk factor for cardiovascular diseases, Zhang et al. discovered that HHcy is an independent risk factor for cardiovascular diseases development, and when combined with HTN may leading to both incidence and overall mortality in Chinese elderly people with no cardiovascular history [29].

Conclusion

Conclusively, the current research indicated that there are strong links between MTHFR gene polymorphism (C677T and A1298C) and the prevalence of hypertension. The two variants were associated with higher odds of HTN in co-dominant, dominant and recessive models with mutant alleles (T and C) playing significant roles in increasing susceptibility to the disease. Also, the patients had significantly higher levels of serum Hcy in comparison with healthy individuals among all the genotypes, indicating that HTN is strongly positively correlated with these genetic variants, HHcy and HTN risk.

Declarations

Consent for publication

This Manuscript has been read by all authors and agreed to be published.

Competing interests

The authors declare that they have no competing interests.

Funding

There are no funding sources for this study.

Author Contribution

Salih M. Al-Khafaji: Conceptualization, Investigation, Writing – original draft, Review & editing, Resources. Visualization. Anwar Madlool Al-janabi: Conceptualization, Writing – original draft, Review & editing, Visualization. Dawood Ali Salim Dawood: Conceptualization, Methodology, Investigation, Data curation and Formal analysis, Resources, Writing – original draft, Review & editing, Visualization. Hamid Jaddoa Abbas: Conceptualization, Resources, Visualization.

Acknowledgement

The authors wish to acknowledge all participants in this study and the staff of the clinical chemistry laboratory at the teaching hospital of Al-Sadder Medical City, Al-Najaf Province, Iraq, for their assistance during sample collection. The authors wish to express their great thanks to Dr. Ali Al-Fahham for his support throughout this work.

Availability of Data and Materials

Data sets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

References

1. Liu F, Lu X *Hypertension and human health: Evidence and prospects*. 2024, Chinese Medical Journals Publishing House Co., Ltd. 42 Dongsi Xidajie … pp. 89–91

2. Muna A, Al-Badri HJ, Mousa NA (2022) Hypertension control among adult Iraqis. J Fac Med Baghdad 64(3):145–152
3. Yuan D et al (2023) Mechanism of homocysteine-mediated endothelial injury and its consequences for atherosclerosis. Front Cardiovasc Med 9:1109445
4. Newton-Cheh C et al (2009) Genome-wide association study identifies eight loci associated with blood pressure. Nat Genet 41(6):666–676
5. Consortium C et al (2011) Genetic variants in novel pathways influence blood pressure and cardiovascular disease risk. Nature 478(7367):103–109
6. Yang B et al (2014) Associations of MTHFR gene polymorphisms with hypertension and hypertension in pregnancy: a meta-analysis from 114 studies with 15411 cases and 21970 controls. PLoS ONE 9(2):e87497
7. Yang KM et al (2014) Methylenetetrahydrofolate reductase C677T gene polymorphism and essential hypertension: A meta–analysis of 10,415 subjects. Biomedical Rep 2(5):699–708
8. Niu W, You Y, Qi Y (2012) Strong association of methylenetetrahydrofolate reductase gene C677T polymorphism with hypertension and hypertension-in-pregnancy in Chinese: a meta-analysis. J Hum Hypertens 26(4):259–267
9. Ward M et al (2020) Impact of the common MTHFR 677C→ T polymorphism on blood pressure in adulthood and role of riboflavin in modifying the genetic risk of hypertension: evidence from the JINGO project. BMC Med 18(1):318
10. Cheng Z et al (2011) Hyperhomocysteinemia impairs endothelium-derived hyperpolarizing factor–mediated vasorelaxation in transgenic cystathionine beta synthase–deficient mice. Blood J Am Soc Hematol 118(7):1998–2006
11. Wald DS, Law M, Morris JK (2002) Homocysteine and cardiovascular disease: evidence on causality from a meta-analysis. BMJ 325(7374):1202
12. Chaussalet M et al (2004) Homocysteine modulates the proteolytic potential of human vascular endothelial cells. Biochem Biophys Res Commun 316(1):170–176
13. Al-janabi AM, Al-Khafaji SM, Faris SA (2022) Association of methyltetrahydrofolate reductase gene mutation, homocysteine level with semen quality of Iraqi infertile males. Egypt J Med Hum Genet 23(1):51
14. Zappa M et al (2024) Genetics of hypertension: from monogenic analysis to GETomics. J Cardiovasc Dev Disease 11(5):154
15. Rios FJ et al (2023) Impact of environmental factors on hypertension and associated cardiovascular disease. Can J Cardiol 39(9):1229–1243
16. Poznyak AV et al (2022) Hypertension as a risk factor for atherosclerosis: Cardiovascular risk assessment. Front Cardiovasc Med 9:959285
17. Fowdar JY et al (2012) Investigation of homocysteine-pathway-related variants in essential hypertension. Int J Hypertens 2012(1):190923

18. Liu H et al (2011) The association between gene polymorphisms of N5, 10-methylenetetrahydrofolate reductase and essential hypertension in patients of Ningxia Hui nationality. Tianjin Med J 39:1095–1098
19. Gai X et al (2009) Association of MMP-9 gene polymorphisms with atrial fibrillation in hypertensive heart disease patients. Clin Chim Acta 408(1–2):105–109
20. Alghasham A et al (2012) Association of MTHFR C677T and A1298C gene polymorphisms with hypertension. Int J health Sci 6(1):3
21. Nassereddine S et al (2015) Association of methylenetetrahydrofolate reductase gene (C677T) with the risk of hypertension in Morocco. BMC Res Notes 8(1):775
22. Meng H et al (2021) Association between MTHFR polymorphisms and the risk of essential hypertension: an updated meta-analysis. Front Genet 12:698590
23. Xu J-R et al (2022) Correlation between hypertension and high homocysteine levels caused by the MTHFR C677T gene polymorphisms. Asian J Surg 45(4):1039–1040
24. Moczulski D et al (2003) Effects of the C677T and A1298C polymorphisms of the MTHFR gene on the genetic predisposition for diabetic nephropathy. Nephrol Dialysis Transplantation 18(8):1535–1540
25. Al-Khafaji SM et al (2015) Genetic aspect of Iraqi pregnant women with pre-eclampsia. Int J Sci Res 4:1563–1567
26. Kuras S et al (2025) Investigation of the effects of MTHFR gene variations and homocysteine levels in hypertensive patients. North Clin Istanbul, 12(1)
27. Zhao W et al (2022) The interaction of hypertension and homocysteine increases the risk of mortality among middle-aged and older population in the United States. J Hypertens 40(2):254–263
28. Moretti R, Caruso P (2019) The controversial role of homocysteine in neurology: from labs to clinical practice. Int J Mol Sci 20(1):231
29. Zhang Z-Y et al (2021) Homocysteine, hypertension, and risks of cardiovascular events and all-cause death in the Chinese elderly population: a prospective study. J geriatric cardiology: JGC 18(10):796

Figures

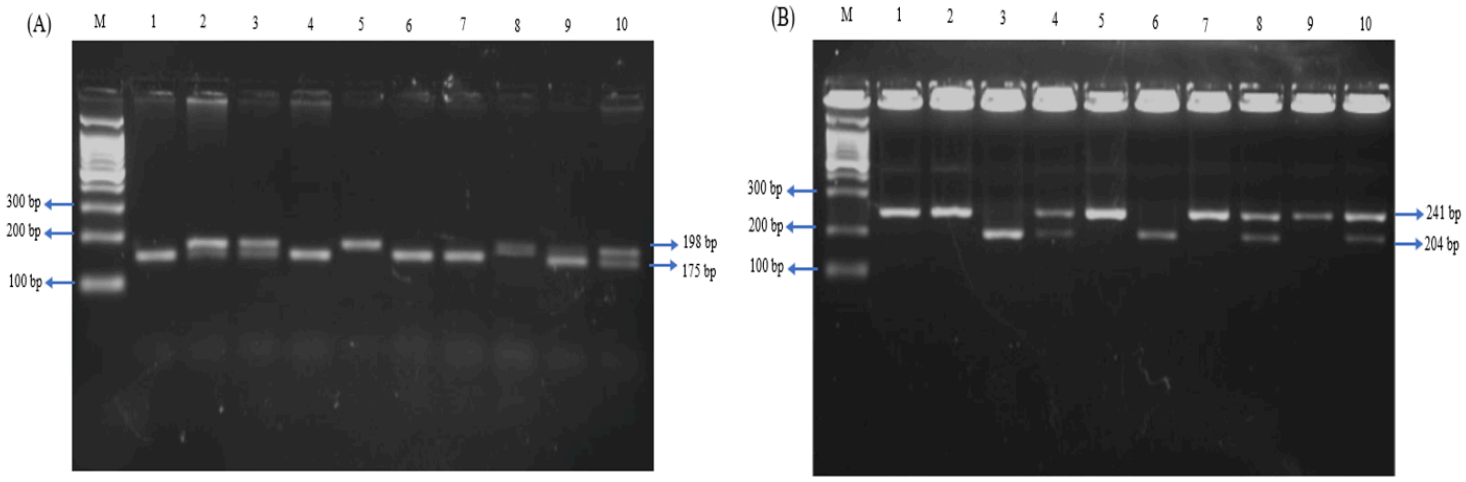


Figure 1

The results of RFLP products. (A) gel electrophoresis of SNP C677T. Lane M indicates 100-bp DNA ladder. Lanes 1, 4, 6 and 7 were genotyped as TT. Lanes 2, 3, 9 and 10 were genotyped as CT. Lanes 5 and 8 were genotyped as CC. (B) gel electrophoresis of SNP A1298C. Lane M indicates 100-bp DNA ladder. Lanes 1, 2, 5, 7 and 9 were genotyped as AA. Lanes 3 and 6 were genotyped as CC. Lanes 4, 8 and 10 were genotyped as AC.