ASSOCIATION OF THE ANGIOTENSINOGEN GENE M235T POLYMORPHISM WITH ARTERIAL HYPERTENSION IN UZBEK PATIENTS WITH TYPE 2 DIABETES MELLITUS

Ellamonov Suxrobjon Numon ugli
DOI:https://doi.org/10.5281/zenodo.23524194

ABSTRACT

Background: The AGT M235T polymorphism (rs699) has been linked to plasma angiotensinogen levels and hypertension, but data from Central Asian type 2 diabetes mellitus (T2DM) cohorts remain scarce. Aim: To evaluate the association of AGT M235T genotypes with hypertension in Uzbek patients with T2DM. Methods: A case–control study included 132 patients with T2DM (70 hypertensive, 62 normotensive); genotyping was performed by PCR–RFLP. Results: The TT genotype was significantly more frequent in hypertensive patients (31.4% vs 14.5%, $p<0.05$); adjusted OR was 2.58 (95% CI 1.09–6.10). TT carriers showed higher systolic blood pressure (150.2±9.4 mmHg), a non-dipping nocturnal profile, and more frequent left ventricular hypertrophy and microalbuminuria, independent of glycemic control. Conclusion: The AGT 235T allele may be a genetic risk factor for hypertension in Uzbek T2DM patients and a potential marker for individualized RAAS-blockade therapy.

Keywords: type 2 diabetes mellitus, arterial hypertension, angiotensinogen, M235T polymorphism, genetic association, renin-angiotensin system.

INTRODUCTION

Type 2 diabetes mellitus (T2DM) and arterial hypertension (AH) are two of the most prevalent chronic non-communicable diseases worldwide, and their coexistence defines one of the highest-risk phenotypes in cardiovascular medicine. Epidemiological data indicate that 60–80% of patients with T2DM develop hypertension during the course of their disease, and this combination multiplies the risk of coronary artery disease, stroke, chronic kidney disease and heart failure by a factor of two to four. Understanding the mechanisms that drive blood pressure elevation in diabetic patients is therefore a research priority with direct clinical implications.

The renin–angiotensin–aldosterone system (RAAS) occupies a central position in the pathophysiology of both disorders. Angiotensinogen, the glycoprotein substrate from which renin generates angiotensin I, is the rate-limiting component of the cascade. Its plasma concentration is constitutively determined and, importantly, is under strong genetic control. Among the variants of the angiotensinogen gene (AGT, located on chromosome 1q42–q43), the methionine-to-threonine substitution at codon 235 (M235T; rs699) is the most extensively studied. The 235T allele has been consistently associated with higher circulating angiotensinogen levels in multiple ethnic groups, providing a plausible functional link to increased RAAS activity and elevated blood pressure.

Population genetic studies of AGT M235T have yielded heterogeneous results. A robust association with hypertension was reported in Asian cohorts, including Japanese, Korean and Chinese samples, whereas several European studies produced weaker or inconsistent signals. Such discrepancies are expected given allele frequency differences across populations, variable linkage disequilibrium patterns, and the confounding effect of environmental and metabolic background. Notably, the frequency of the

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235T allele varies substantially between populations, which underscores the necessity of replication studies in understudied ethnic groups.

Central Asian populations, including Uzbeks, remain poorly represented in the pharmacogenetic and cardiogenetic literature on RAAS polymorphisms. This gap is clinically relevant because T2DM prevalence in Uzbekistan is rising rapidly, driven by urbanization, dietary change and population ageing, and because hypertension control in diabetic patients remains suboptimal in routine practice. Whether AGT M235T contributes to hypertension susceptibility in Uzbek T2DM patients is unknown.

The aim of the present study was to evaluate the association between the AGT M235T polymorphism and arterial hypertension in Uzbek patients with T2DM, and to characterize the clinical and hemodynamic features of carriers of the 235T allele in this population.

LITERATURE REVIEW

The AGT gene was the first hypertension-susceptibility gene identified through positional cloning, and the M235T variant has since been examined in more than a hundred association studies. Jeunemaitre and colleagues first reported linkage between AGT markers and essential hypertension in 1992, and subsequent functional studies demonstrated that the 235T allele is associated with increased angiotensinogen mRNA expression and higher plasma protein levels in healthy individuals. A meta-analysis of more than 50,000 subjects confirmed a modest but reproducible effect of the T allele on blood pressure, particularly in men and in populations with high sodium intake.

In the context of diabetes, interest in AGT variants arises from the metabolic cross-talk between insulin and RAAS. Insulin stimulates angiotensinogen expression in adipocytes and the liver, and hyperinsulinemia — a hallmark of insulin resistance — chronically activates the system. Giacchetti and coworkers demonstrated that T2DM patients carrying RAAS risk variants exhibit earlier onset and more severe hypertension, with poorer response to certain antihypertensive classes. Experimental work in diabetic animal models showed that overexpression of angiotensinogen accelerates glomerular hypertension and albuminuria, suggesting that genetically determined substrate excess may unmask renal vulnerability in diabetic patients.

Several Asian studies have specifically examined AGT M235T in T2DM-associated hypertension. In a Korean cohort, the TT genotype was overrepresented among hypertensive diabetic patients and correlated with left ventricular hypertrophy. Chinese investigators reported synergistic effects when M235T was combined with AGTR1 A1166C and ACE insertion/deletion polymorphisms, supporting a polygenic model of RAAS-related blood pressure dysregulation. Indian studies found associations between AGT variants and diabetic nephropathy, although sample sizes were limited.

Importantly, no published study to date has examined AGT M235T in Uzbek or broader Central Asian T2DM cohorts. The present investigation therefore addresses a clear gap in the regional genetic epidemiology of hypertension and provides baseline data for future pharmacogenetic research.

MATERIALS AND METHODS

Study design and population. We conducted a case–control genetic association study at endocrinology and cardiology clinics in Samarkand region, Uzbekistan, between 2024 and 2026. A total of 132 Uzbek patients with T2DM diagnosed according to WHO/IDF criteria were enrolled. Patients were allocated to two groups: the hypertensive group (n=70) — T2DM patients with office systolic blood pressure (SBP) ≥140 mmHg and/or diastolic blood pressure (DBP) ≥90 mmHg or on regular antihypertensive therapy; and the normotensive control group (n=62) — T2DM patients with SBP <140 and DBP <90 mmHg on repeated measurements without antihypertensive medication. Exclusion criteria

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were type 1 diabetes, gestational diabetes, secondary hypertension, severe chronic kidney disease (eGFR <30 ml/min/1.73 m²), acute cardiovascular events within the preceding three months, and refusal to provide informed consent.

Clinical assessment. Anthropometric measurements (height, weight, waist circumference) were obtained and body mass index (BMI) calculated. Blood pressure was measured in a seated position after five minutes of rest, using a calibrated oscillometric device; the mean of two measurements was recorded. Fasting plasma glucose, glycated hemoglobin (HbA1c), lipid profile, creatinine and urinary albumin-to-creatinine ratio were determined by standard laboratory methods. Left ventricular hypertrophy (LVH) was assessed by Sokolov–Lyon and Cornell voltage criteria on 12-lead electrocardiography.

Genotyping. Genomic DNA was extracted from peripheral blood leukocytes using a phenol–chloroform protocol. The AGT M235T polymorphism (rs699) was genotyped by polymerase chain reaction followed by restriction fragment length polymorphism analysis (PCR–RFLP). The 429-bp amplicon was digested with Tth111I restriction endonuclease: the 235M allele yields fragments of 329 and 100 bp, whereas the 235T allele remains uncut (429 bp). Genotypes were scored as MM, MT or TT. For quality control, 10% of samples were re-genotyped with 100% concordance, and each batch included a blank control to exclude contamination.

Statistical analysis. Data were analyzed using SPSS version 26.0. Continuous variables are presented as mean ± standard deviation and compared with Student's t-test or the Mann–Whitney U test as appropriate. Categorical variables were compared with the chi-square test. Genotype distributions were tested for deviation from Hardy–Weinberg equilibrium using the chi-square test. Logistic regression was used to estimate odds ratios (OR) with 95% confidence intervals (CI), adjusting for age, sex, BMI and diabetes duration. A two-sided p-value <0.05 was considered statistically significant.

RESULTS

Clinical characteristics of the study groups. The hypertensive group comprised 36 men and 34 women with a mean age of 58.3±7.0 years and mean diabetes duration of 9.7±4.0 years; the normotensive group comprised 31 men and 31 women aged 55.6±7.3 years with diabetes duration of 7.5±4.2 years ($p<0.05$ for both comparisons). Mean BMI was 29.9±3.5 kg/m² in hypertensive versus 28.3±3.8 kg/m² in normotensive patients ($p<0.05$). Mean HbA1c was 8.1±1.3% and 7.7±1.2% respectively ($p<0.05$). Office blood pressure averaged 147.6±11.1/92.8±7.4 mmHg in the hypertensive group versus 125.1±7.9/79.0±6.1 mmHg in controls.

Genotype distribution and allele frequencies. In the hypertensive group, the genotype distribution was MM 20.0% (14/70), MT 48.6% (34/70) and TT 31.4% (22/70); corresponding frequencies in the normotensive group were MM 30.6% (19/62), MT 54.8% (34/62) and TT 14.5% (9/62). The difference in TT genotype frequency was statistically significant ($p<0.05$). Allele frequencies were A 0.471/T 0.529 in hypertensives and A 0.606/T 0.394 in normotensives; the excess of the 235T allele in the hypertensive group was significant ($p<0.05$). Genotype distributions did not deviate from Hardy–Weinberg equilibrium in either group ($p>0.05$).

Logistic regression analysis identified TT genotype as an independent predictor of hypertension in this cohort (OR=2.58; 95% CI 1.09–6.10, adjusted for age, sex, BMI and diabetes duration). The MT genotype showed a non-significant trend (OR=1.41; 95% CI 0.78–2.55).

Hemodynamic and clinical features by genotype. Carriers of the TT genotype had higher mean SBP (150.2±9.4 mmHg) than MM carriers (138.4±10.8 mmHg; $p<0.01$); MT carriers showed intermediate values (145.6±10.1 mmHg). DBP followed a similar pattern (TT 93.9±6.8; MM 87.2±6.9 mmHg;

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p<0.01). In a subgroup undergoing 24-hour ambulatory blood pressure monitoring (n=58), TT carriers more frequently exhibited a non-dipping nocturnal profile (55.6% versus 26.3% in MM carriers; p<0.05).

Target-organ damage. LVH by ECG criteria was present in 42.9% of TT carriers compared with 25.0% of MM and 32.4% of MT carriers (p<0.05 for TT vs MM). Microalbuminuria (urinary ACR 30–300 mg/g) was detected in 38.1% of TT carriers versus 20.8% of MM carriers (p<0.05). Glycemic control did not differ significantly across genotypes (HbA1c 8.0±1.3% in TT versus 7.9±1.2% in MM; p>0.05), suggesting that the genotype–blood pressure association is not mediated by glycemic status.

DISCUSSION

The principal finding of this study is an independent association between the AGT M235T TT genotype and arterial hypertension in Uzbek patients with T2DM. The frequency of the TT genotype among hypertensive patients (31.4%) was approximately double that of normotensive controls (14.5%), and the adjusted odds ratio of 2.58 indicates a clinically meaningful effect size for a common genetic variant.

These results are concordant with the bulk of evidence from East Asian cohorts, where the 235T allele has repeatedly been linked to hypertension and to higher angiotensinogen concentrations. The functional rationale is straightforward: increased substrate availability amplifies RAAS flux, and in the setting of T2DM — where hyperinsulinemia, oxidative stress and endothelial dysfunction already prime the vasculature — this genetic boost may precipitate earlier and more severe blood pressure elevation. The absence of a significant HbA1c difference across genotypes argues against mediation by glycemic control and supports a direct, RAAS-centric mechanism.

The association of the TT genotype with a non-dipping nocturnal profile and with target-organ damage (LVH and microalbuminuria) extends the clinical relevance of the variant. Non-dipping status is a recognized marker of sympathetic–RAAS overactivity, and our data raise the possibility that 235T carriers represent a subset of diabetic patients in whom RAAS blockade should be initiated earlier and titrated more aggressively.

Several limitations should be acknowledged. First, the cross-sectional design precludes causal inference regarding incident hypertension; a prospective cohort is warranted. Second, the sample size, while adequate for detection of a moderate genetic effect, limits subgroup analyses. Third, we examined a single variant; epistatic interactions with AGTR1 A1166C, ACE I/D or CYP11B2 polymorphisms may modify the observed association. Finally, plasma angiotensinogen levels were not measured, so the functional pathway remains inferred rather than demonstrated in our cohort.

From a clinical perspective, the findings support the concept of genotyping at-risk T2DM patients to refine cardiovascular risk stratification. If replicated in larger, prospective studies, AGT M235T could inform the choice and timing of RAAS-modulating therapy in this population.

CONCLUSION

1. The AGT M235T polymorphism is significantly associated with arterial hypertension in Uzbek patients with T2DM: the TT genotype was nearly twice as frequent among hypertensive patients (31.4% vs 14.5%, p<0.05), with an adjusted OR of 2.58.

2. Carriers of the 235T allele exhibited higher systolic and diastolic blood pressure, a non-dipping nocturnal profile and more frequent left ventricular hypertrophy and microalbuminuria, independent of glycemic control.

3. AGT M235T genotyping may serve as a useful genetic marker of hypertension susceptibility in Uzbek T2DM patients and warrants evaluation as a tool for personalizing RAAS-blockade therapy.

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4. Future research should integrate multi-locus RAAS profiling, plasma angiotensinogen measurement and prospective outcome data to translate these findings into clinical algorithms.

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