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Gender-specific Association of K173R-CYP11B2 Polymorphism with Type 2 Diabetes Mellitus in Moroccan Population

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Abstract

Aldosterone Synthase (CYP11B2) is a critical enzyme involved in the synthesis of aldosterone, and polymorphisms within its gene have been proposed to play a significant role in the pathogenesis of Type 2 Diabetes Mellitus (T2DM). This study aimed to investigate the association between the K173R polymorphism in the CYP11B2 gene and gender within a Moroccan population with T2DM. A total of 90 women (T2DM=57, Control=33) and 110 men (T2DM=43, Control=67) from the Moroccan population were included in the study. Polymerase Chain Reaction Restriction Fragment Length Polymorphism (PCR-RFLP) was utilized to identify the CYP11B2 (K173R) polymorphism. Results revealed that the distribution of KK, KR, and RR genotypes in women with T2DM were 38.6%, 47.4%, and 14.00%, respectively, compared to 55.8%, 34.9%, and 9.3% in the control women group. Similarly, in men with T2DM, the frequencies were 44.2%, 44.2%, and 11.6%, while in control men, they were 61.4%, 28.1%, and 10.5%, respectively. Allele frequencies showed that K and R alleles comprised 62.28% and 37.72% in women with T2DM, compared to 73.25% and 26.75% in control women, and 66.28% and 33.72% in men with T2DM, compared to 71.64% and 28.36% in control men, respectively. No significant differences in genotypic and allelic frequencies were observed between genders. In conclusion, our study indicates no association between the CYP11B2 (K1734) polymorphism and gender within our Moroccan population sample.

Keywords: Type 2 diabetes mellitus; CYP11B2; K173R polymorphism; Gender; Morocco

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1. Introduction

The diabetes mellitus (DM) is a complex metabolic disease affecting more than 10% of the global population. Over 90% of diabetes mellitus cases are type 2 diabetes mellitus (T2DM) [1]. Besides its direct health implications, T2DM is also a major contributor to coronary heart disease, strokes, peripheral vascular disease, end stage renal disease, retinopathy, neuropathy, and lower limb amputations [2]. While the prevalence of diabetes is similar between genders, significant disparities exist in how the disease manifests and progresses based on gender [3]. Global epidemiological data indicate that adult women tend to receive a diagnosis of T2DM later in life, often at a stage where the disease has already progressed further [3]. Additionally, diabetic women are more likely to develop cardiovascular complications and have a higher risk of diabetes related deaths than their men counterparts [4]. These observed gender differences underscore the importance of better understanding the impact of sex and gender on metabolic risk associated with diabetes. Recent research suggests that these disparities may be influenced by genetic factors [5–7], among which the K173R polymorphism of the CYP11B2 gene is implicated [8–10]. This gene is involved in aldosterone production, a hormone that regulates sodium reabsorption and potassium excretion in the kidneys. Although this polymorphism has been associated with cardiovascular conditions such as hypertension, its role in the development of T2DM remains relatively understudied [11]. This research gap motivates our retrospective case-control study, which aims to evaluate the distribution of the K173R polymorphism among T2DM patients and healthy controls, with a particular focus on its interaction with gender within the Moroccan population. By better understanding how genetic variations interact with gender specific risk factors, we hope to identify new strategies for preventing and treating T2DM more effectively, especially in women and gender minorities.

2. Materials and Methods

2.1. Study Participants

A total of 161 participants were enrolled in this study, comprising 86 patients diagnosed with T2DM and 75 control subjects. Participants were recruited from individuals consulting at the Diagnostic Center-Moulay Youssef Hospital in Rabat. Blood samples were collected during routine tests requested by their attending physicians, with 1ml of blood retained from each participant for genomic DNA extraction. Ethical approval was obtained, and all participants provided informed consent in accordance with the principles outlined in the Declaration of Helsinki.

2.2. Patient Selection and Control Group

Patients diagnosed with T2DM were identified through their medical records, with diagnoses confirmed by attending physicians. The control group comprised individuals without a history of T2DM or other relevant medical conditions, selected from the same population pool as the patients to ensure demographic comparability. Matching of the control group with the patient group based on age and gender was performed to reduce potential confounding variables.

2.3. Genomic DNA Extraction and Genotyping

Genomic DNA was extracted from blood samples using the high ionic protein recharging method [12]. The K173R polymorphism of the CYP11B2 gene was determined using polymerase chain reaction (PCR) Restriction Fragment Length Polymorphism (RFLP). A 371 bp fragment was amplified using specific primers, and PCR products were digested with the Bsu36I restriction enzyme. Genotyping was determined based on the presence or absence of the restriction site due to the substitution of arginine (R) by lysine (K) at position 173. Fragment analysis was performed using electrophoresis on 2% agarose gels.

2.4. Statistical Analysis

Data analysis was performed using IBM SPSS Statistics 24 software package. Descriptive statistics were computed to determine means and standard deviations of variables. The significance of genotype distribution differences between patient and control groups was assessed using the χ^2 statistics test. Analysis of basic clinical characteristics and genotype/allelic frequencies by gender was conducted. Odds ratios (OR) with 95% confidence intervals (CI) were computed to estimate diabetes risk associated with various genotype combinations. Statistical significance was set at $p \leq 0.05$.

3. Results and discussion

Table 1 provides a comprehensive overview of the baseline clinical characteristics of both men and women with type 2 diabetes and healthy controls. Notably, significant differences were observed solely in waist circumference and SBP between control men and women ($p<0.05$). Conversely, there were no statistically significant variations noted in age, glycemia, TC, HDL, LDL, and TG between men and women with T2DM and their respective control groups. The distribution of the K173R polymorphism within the CYP11B2 gene, categorized by gender, is delineated in Table 2, inclusive of allele frequencies among individuals diagnosed with T2DM and healthy controls. In women with T2DM, the frequencies of the KK, KR, and RR genotypes were 38.6%, 47.4%, and 14.00%, respectively, while in control women, these frequencies were 55.8%, 34.9%, and 9.3%. Notably, genotype distributions of the CYP11B2 polymorphism differed marginally between men with T2DM and control women, though the disparity did not achieve statistical significance ($\chi^2 = 2.946$, $p = 0.229$). For men with T2DM, the KK, KR, and RR genotypes accounted for 44.2%, 44.2%, and 11.6% of the subjects, respectively, whereas in men controls, these frequencies were 61.4%, 28.1%, and 10.5%. Similarly, no statistically significant difference in genotype distribution was observed between men with T2DM and women controls ($\chi^2 = 0.718$, $p = 0.698$), indicating comparable distributions across both groups. Examining allele frequencies, the R allele displayed a slightly higher prevalence among women T2DM subjects (37.72%) compared to male controls (26.75%), whereas the K allele exhibited greater prevalence among women controls (73.25%) than among women T2DM subjects (26.75%). Among men participants, the R allele was marginally more prevalent among those with T2DM (33.72%) than among controls (28.36%), whereas the K allele was more abundant among men controls (71.64%) compared to men T2DM subjects (66.28%). This study represents the first endeavor to explore the gender specific correlation between the K173R polymorphism of the aldosterone synthase gene and T2DM within the Moroccan population. Employing a case-control methodology, our investigation yielded no statistically significant differences in the genotype and allele frequencies of the CYP11B2 (K173R) polymorphism between men and women among both individuals with T2DM and healthy controls ($p>0.05$). The distribution of the K173R polymorphism within the CYP11B2 gene exhibits notable variation across different populations. European populations demonstrate a nearly equal distribution of the K and R alleles, with K occurring in 52% and R in 48% of individuals [10]. Conversely, populations with African heritage show a significantly higher prevalence of the K allele, reaching 81%. This trend extends to Iberian populations in Spain, where the frequency of the KK gene is elevated by 79%. This finding is also consistent with the predominance of the KK gene among individuals of African descent outside of Africa [10]. The K173R polymorphism of the CYP11B2 gene has been implicated in various metabolic conditions, including plasma glucose levels, glucose intolerance, and T2DM [13]. Although the K173R polymorphism may not have direct functional implications on its own, it may be linked to other functional polymorphisms, such as -344T/C, which has been associated with T2DM [8,14–17]. Previous studies have identified a strong linkage disequilibrium between K173R and -344C/T [9,18,19], suggesting potential interactions between these genetic variants that could provide insights into the complex genetic basis of diabetes.

Table 1. Anthropometric and metabolic characteristics of the study population

Parameters	T2DM case		P-value	Control		P-value
	Men	Women		Men	Women	
Age (Year)	52.79 ± 8.50	54.47 ± 11.53	0.423	48.86 ± 12.62	48.86 ± 12.62	0.757
Glycemia (g/l)	1.95 ± 0.68	1.91 ± 0.58	0.691	0.88 ± 0.09	0.88 ± 0.88	0.826
Waist (cm)	90.32 ± 6.68	88.63 ± 8.08	0.267	83.63 ± 6.27	80.83 ± 5.42	0.022
TC (g/l)	1.98 ± 0.55	1.95 ± 0.39	0.740	1.79 ± 0.42	1.77 ± 0.36	0.839
HDL (g/l)	0.57 ± 0.29	0.49 ± 0.18	0.113	0.57 ± 0.19	0.59 ± 0.16	0.713
LDL (g/l)	1.10 ± 0.44	1.14 ± 0.41	0.650	1.04 ± 0.38	1.01 ± 0.31	0.645
TG (g/l)	1.53 ± 0.94	1.55 ±1.07	0.933	0.83 ± 0.68	0.86 ± 0.33	0.648
SBP (mmHg)	142.13 ± 20.43	137.59 ± 14.23	0.193	130.08 ± 10.97	125.74 ± 6.33	0.022
DBP (mmHg)	79.13 ± 11.12	75.43 ± 9.20	0.072	72.38 ± 6.10	70.88 ± 4.29	0.172

The data are presented in Forms: Mean±SD. SD. Standard Deviation; T2DM. Type 2 Diabetes Mellitus; WC. Waist Circumference; SBP. Systolic Blood Pressure; DBP. Diastolic Blood Pressure; TG. TriGlyceride; TC. Total Cholesterol; LDL. Low-Density Lipoprotein Cholesterol; HDL. High-Density Lipoprotein Cholesterol.

Table 2. The CYP11B2 (-344 C/T) polymorphism genotype and allele frequencies in T2D patients and control group distributed according to gender

Genotypes/Alleles	Women				Men			
	T2DM case	Control	OR (95% CI)	P-value	T2DM case	Control	OR (95% CI)	P-value
Genotypes								
KK	22 (38.6%)	24 (55.8%)	0.49 (0.22-1.11)	0.089	19 (44.2%)	35 (61.4%)	0.72 (0.33-1.56)	0.410
KR	27 (47.4%)	15 (34.9%)	1.68 (0.74-3.79)	0.212	19 (44.2%)	26 (28.1%)	1.24 (0.57-2.71)	0.560
RR	8 (14.0%)	4 (9.3%)	1.5 9(0.44-5.67)	0.473	5 (11.6%)	6 (10.5%)	1.33 (0.38-4.68)	0.649
Alleles								
K	71 (62.28%)	63 (73.25%)			57 (66.28%)	96 (71.64%)		
R	43 (37.72%)	23 (26.75%)	1.65 (0.90-3.05)	0.103	29 (33.72%)	38 (28.36%)	1.28 (0.71-2.30)	0.400

T2DM. Type 2 Diabetes Mellitus; **OR.** Odd Ratio; **CI.** Confidence Interval.

While investigating the association of genetic polymorphisms of CYP11B2 with Metabolic Syndrome (MetS) in a Korean population, Kim et al. did not find a strong association between the K137R polymorphism of CYP11B2 and MetS. Additionally, Ranade et al. observed a significant correlation between fasting plasma glucose levels and the K173R polymorphism, with individuals homozygous for the R allele exhibiting higher mean fasting glucose levels than those with KR or RR genotypes [8]. This observation suggests that the K173R polymorphism may influence the expression or activity of the enzyme, thereby affecting aldosterone levels. Alternatively, the R allele at position 173 may diminish aldosterone synthase activity, resulting in a slower conversion of an intermediary substance to aldosterone. This could lead to a slight accumulation of intermediary compounds, such as corticosterone, a mild glucocorticoid, aligning with the recessive impact of the arginine allele on glucose levels [8].

4. Conclusions

In conclusion, our study sheds light on the distribution and potential implications of the K173R polymorphism in the Moroccan population, contributing to our understanding of the genetic factors underlying T2DM susceptibility. Further research is warranted to elucidate the intricate interactions between genetic variants and metabolic conditions and to validate these findings in diverse populations.

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