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Research Article

ROLE OF ARYL HYDROCARBON RECEPTOR NUCLEAR TRANSLOCATOR IN DIABETES MELLITUS AMONG RAICA AND NON-RAICA COMMUNITIES OF RAJASTHAN

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ABSTRACT

Objective: This study was conceptualized to assess genotypic factors associated with a lower prevalence of type-2 diabetes mellitus in Raica community of Rajasthan, India.

Methods: In this study, 114 people from Raica community and 150 people from non-Raica community were recruited. Their demographic details age and sex, anthropometric data body mass index, and waist-to-hip ratio, and laboratory parameters such as fasting blood glucose (FBG), HbA1c, and physiological parameters of systolic and diastolic blood pressure were taken into consideration.

Results: In the present study, there were 40 females and 74 males from Raica community and, 52 females and 98 males from non-Raica community. The mean age was 41.14 and 46.93 in Raica and non-Raica communities. The FBG, HbA1c levels, and physiological parameters were significantly lower in Raica community ($p<0.05$). The AG allele of the Aryl hydrocarbon receptor nuclear translocator gene was more frequently seen in individuals with lower FBG and no individual studied, had the AA allele.

Conclusion: The genetic polymorphism studied has the same frequency of distribution in both Raica and non-Raica people with or without diabetic conditions. The leanness and better control over the glucose levels in the Raica community are supposed to be key factors in lowering diabetes mellitus prevalence.

Keywords: ARNT gene, Camel milk, Raica community, Type-2 diabetes.

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INTRODUCTION

Diabetes mellitus is a group of metabolic diseases whose common feature is hyperglycemia caused by defective insulin secretion or by a defective tissue response to insulin. Diabetes mellitus is one of the most common and fastest-growing metabolic diseases worldwide. As per the World Health Organization, there are more than 422 million people living with diabetes mellitus by 2014. Prevalence has been rising more rapidly in middle-and low-income countries [1].

The prevalence of diabetes mellitus varies in different geographical regions, ethnic groups, and age groups. Lifestyle factors such as sedentary life, smoking, obesity, and dietary factors [2]. The first authentic data on the prevalence of diabetes in rural and urban India came from the multicentric study conducted by the India Council of Medical Research, India, in the early 1970s, which reported a prevalence of 2.3% and 1.5% in urban and rural populations, respectively [3]. Later on, similar multicentric studies also showed a higher prevalence in urban populations [4-6].

Despite known roles for obesity, sedentary lifestyles, and diet, genetic predisposition accounts for a significant risk for diabetes mellitus. Various genes currently considered to be associated with diabetes mellitus susceptibility include those encoding for insulin, insulin receptor, insulin receptor substrate, prohormone convertase, glucokinase, glucose transporters, glucose-dependent insulinotropic polypeptide receptor, glycogen synthase, cysteine protease calpain 10, leptin gene (Ob), apolipoprotein E, peroxisome proliferator-activated receptor gamma, and Aryl hydrocarbon receptor nuclear translocator gene (ARNT). Unfortunately, very little data are available on diabetes mellitus susceptibility loci in the Asian Indian population. Moreover, variations in disease prevalence among ethnic groups that share a similar environment suggest a strong genetic contribution to disease predisposition [6,7].

In our region, a Raica community is known to have a low prevalence of diabetes mellitus. The present study was conceptualized to know the genetic factors associated with diabetes mellitus. In this study, we compared Raica and non-Raica community participants for ANRT G1511A genotype distribution to know the difference in diabetes mellitus prevalence.

METHODS

This was a case–control study. Samples and clinical parameters of selected patients were compared with those of age and sex-matched healthy adults.

Department of Biochemistry and Diabetes Care and Research Centre, Department of Medicine, S.P. Medical College, Bikaner, conducted this study jointly. Patients with type-2 diabetes were recruited into the study. Patients who had comorbid conditions such as renal failure, hepatic failure, and coronary heart disease and pregnant females were excluded from the study. Demographic, biochemical, and anthropologic parameters essential for the study were recorded.

Blood samples were collected from participants. Samples were subjected to DNA isolation, quantification of DNA, and polymerase chain reaction (PCR). The forward and reverse primers for PCR amplification were *ARNT-511F 5'-TATTTGTCTTGTCAACTGGCCTTT-GAC-3'* and *ARNT-511R 5'-CTGGCCAGTCCTCTCTTCCTGGG-AC-3*. Amplified products were subjected to RFLP for detection of ARNT polymorphism.

Ethical statement

Patient consent was taken before starting the study. This study was approved by the Rajasthan University of Health Sciences (Research Section). Letter No. F-7/Research/RUHS/2007-08/5675 date: January 29, 2008.

RESULTS

In the present study, a total of 114 participants from Raica community and 150 participants from non-Raica community were recruited to estimate the distribution of ARNT gene alleles in both these communities.

Among Raica and non-Raica communities, a significant difference ($p<0.05$) was observed in different anthropometric and clinical parameters. The fasting blood glucose (FBG) levels and systolic as well as diastolic blood pressure of Raica people were lower in comparison to non-Raica people (Table 1).

The frequency of distribution of GG, AG, and AA alleles of ARNT genes revealed that the allelic distribution in both communities was not significant. The allele distribution for both communities satisfies Hardy Weinberg equilibrium ($p>0.5$) (Table 2).

The difference in genotype and allelic distribution in diabetics and non-diabetics of Raica and non-Raica communities showed GG allele seems to be associated with higher FBG levels. The AG allele is more frequently seen in individuals with lower FBG. It is noteworthy that no individuals had the AA allele. There were three heterozygous individuals in <23 kg/m^2 body mass index (BMI) group in Raica residents, whereas only one heterozygous individual was found in the non-Raica group. Even though the distribution is not statistically significant because of the small numbers, the odds ratio was 1.645 with a wide confidence interval. In the Raica community, there is an almost equal distribution of individuals in <0.85 and >0.85 waist-to-hip ratio (WHR) groups, while three heterozygous individuals fall in the >0.85 WHR group among Raica members. The odds ratio of 2.067 seems a little large but is still not supported by the corresponding community, where there is an almost equal distribution of confidence intervals, which shows that pieces of evidence are yet to be fully explored in a larger cohort (Table 3).

Table 1: Comparison of demographic profile, clinical, and anthropometric parameters between Raica and non-Raica community

Participants' characteristics	Ethnicity		p-value
	Raica community (n=114) (Mean±SD)	Non-Raica community (n=150) (Mean±SD)	
Age	41.14±17.92	46.93±12.37	>0.05
Body mass index	18.97±3.12	24.97±6.10	<0.0001
Fasting blood glucose	91.39±14.16	147.47±67.20	<0.0001
Waist-hip ratio	0.84±0.09	0.92±0.13	<0.0001
Hba1c	5.05±0.59	7.79±1.97	<0.0001
Systolic blood pressure	119.66±19.31	124.17±12.5	>0.05
Diastolic blood pressure	75.55±10.98	82.92±9.37	<0.05

$p<0.05$ was considered significant. SD: Standard deviation

Table 2: Distribution of ARNT G1511A genotype and allele type in males and females of Raica and non-Raica community

Participants	ARNT G1511A genotype											
	Raica community (n=114)						Non-Raica community (n=150)					
	N	GG	AG	AA	G	A	N	GG	AG	AA	G	A
Female	40	38 (0.95)	02 (0.05)	00 (0.00)	78 (0.97)	02 (0.03)	52	50 (0.96)	02 (0.04)	00 (0.00)	102 (0.98)	02 (0.02)
Male	74	68 (0.91)	06 (0.09)	00 (0.00)	142 (0.95)	06 (0.05)	98	92 (0.93)	06 (0.07)	00 (0.00)	190 (0.96)	06 (0.02)
Total	114	106 (0.92)	08 (0.08)	00 (0.00)	220 (0.96)	08 (0.04)	150	142 (0.94)	08 (0.06)	00 (0.00)	292 (0.97)	08 (0.03)

Total
Odds ratio=1.327, CI (95%)=0.355–4.957
Hardy Weinberg Equilibrium, p=0.783670 (Raica) and 0.812449 (non-Raica)
Female
Odds ratio=1.308, CI (95%)=0.132–12.913
Hardy Weinberg Equilibrium, p=0.908707 (Raica) and 0.920360 (non-Raica)
Male
Odds ratio=1.338, CI (95%)=0.299–5.981
Hardy Weinberg Equilibrium, p=0.797165 (Raica) and 0.825051 (non-Raica)
ARNT: Aryl hydrocarbon receptor nuclear translocator

Table 3: Distribution of ARNT G1511A genotype and allele type in subjects of Raica and non-Raica communities according to clinical parameters

Participants' clinical characteristics	ARNT G1511A genotype											
	Raica community (n=114)						Non-Raica community (n=150)					
	N	GG	AG	AA	G	A	N	GG	AG	AA	G	A
FBG >126mg/dl	04	04 (0.92)	00 (0.00)	00 (0.00)	08 (1.00)	00 (0.00)	92	84 (0.91)	08 (0.09)	00 (0.00)	176 (0.95)	08 (0.05)
FBG <126MG/dl	110	102 (0.92)	08 (0.08)	00 (0.00)	212 (0.96)	08 (0.04)	58	58 (1.00)	00 (0.00)	00 (0.00)	116 (0.00)	00 (0.00)
BMI >23	18	16 (0.88)	02 (0.12)	00 (0.00)	34 (0.94)	02 (0.06)	98	92 (0.93)	06 (0.07)	00 (0.00)	190 (0.96)	06 (0.04)
BMI <23	96	90 (0.93)	06 (0.07)	00 (0.00)	186 (0.95)	06 (0.05)	52	50 (0.93)	02 (0.04)	00 (0.00)	102 (0.98)	02 (0.02)
WHR >0.85	48	42 (0.87)	06 (0.13)	00 (0.00)	90 (0.93)	06 (0.07)	32	30 (0.93)	02 (0.07)	00 (0.00)	62 (0.96)	02 (0.04)
WHR <0.85	66	64 (0.96)	02 (0.04)	00 (0.00)	130 (0.00)	02 (0.98)	118	112 (0.94)	06 (0.06)	00 (0.00)	230 (0.97)	06 (0.03)

FBG: No statistical analysis is possible as one rank contains 00 as a value. BMI <23 Odds ratio=1.65, CI (95%)=0.228–11.722, WHR >0.85 Odds ratio=2.067, CI (95%)=0.278–14.970, FBG: Fasting blood glucose; BMI: Body mass index; WHR: Waist-to-hip ratio

DISCUSSION

The prevalence of diabetes among people between the ages of 20 and 70 is projected to be 8.7%, making it an increasing problem in India. Around 422 million people worldwide had diabetes as of 2014. An estimated 2 million people died from diabetes worldwide in 2019 [1].

By region and ethnicity, type-2 diabetes prevalence varies substantially. When exposed to an environment with an abundance of food and little physical activity, type-2 diabetes is viewed as an illustrative example of a multifactorial polygenic disease in which common genes interact to produce the condition [8,9].

One hundred and fifty participants from non-Raica groups and 114 members of the Raica community participated in the current study. The average age of Raica people was 41.14±17.92, compared to 46.93±12.37 for non-Raica people. The Raica community members had greater control over the anthropometric and clinical characteristics that are indicators of diabetes risk. A highly significant difference in the distribution of BMI, FBS, WHR, HbA1c ($p<0.001$), and diastolic BP significant ($p<0.05$) was observed. The frequency of GG, AG, and AA allele of the ARNT gene was studied using PCR-RFLP method. The frequency of GG allele in Raica community was 0.92 and in non-Raica community was 0.94 and the difference was not significant. Similarly, the distribution of AG allele in Raica community was 0.08, and in non-Raica community was 0.06 and the difference was statistically insignificant. A similar finding was reported by Cao and Hegele among Caucasians and Africans where the distribution of GG allele and AG allele was almost similar as no AA allele was reported by them but the difference of BMI, FBS, WHR, and HbA1c is more significant in our study than study conducted by Cao and Hegele [10].

There were no discernible differences in the genotype and allele distribution between the Raica and non-Raica. The data were subsequently divided into groups according to various clinical and anthropometric criteria, and the distribution of genotypes was examined. The odds ratio in our analysis for the WHR was significant (2.067), but the confidence interval did not support it because there was a complete lack of allele A.

The distribution of ARNT G1511A genotype and allele type in subjects of Raica and non-Raica community with fasting blood sugar was compared. In Raica community, 97.5% of subjects had fasting blood sugar levels below 126 mg/dL compared to non-Raica where only 38.6% of individuals had a fasting blood sugar below 126 mg/dL. This observation once again confirms the low incidence of type-2 diabetes mellitus among Raica subjects. However, statistical analysis was not possible because only two reported cases of diabetes in Raica community have been found [10,11]. In our study, it was concluded that GG allele seems to be associated with higher FBG levels. The AG allele is more frequently seen in individuals with lower FBG. It is noteworthy that no individual studied had the AA allele.

In this study, we have also compared the genotype and allelic distribution with BMI in Raica and non-Raica communities. There are heterozygous individuals in <23 kg/m^2 BMI group in Raica residents whereas only one heterozygous individual was found in the non-Raica group. Even though the distribution is not statistically significant because of the small numbers still the odds ratio was 1.645 with a wide confidence interval.

We have also compared the genotype and allelic distribution of both communities with WHR. In Raica community, the frequency of WHR (0.85) in GG allele was 0.87, and in AG allele was 0.13 compared to non-Raica community in which it was 0.93 with GG allele and 0.07 with AG allele. The frequency of WHR (<0.85) with GG allele in Raica community was 0.96 and with AG allele in the same community was 0.04. As opposed to this in non-Raica community the frequency for WHR (<0.85) for GG allele was 0.94 and for AG allele was 0.06. Again the difference was found to be significant. The findings are supported by other studies. This could explain the negligible incidence of diabetes mellitus in Raica community but for confirmed evidence, it has to be explored in a large cohort. Apart from genotypic differences, Raica community consumes camel milk which has been found to show an alternate treatment regimen by some authors [12-14].

CONCLUSION

The present study concluded that the polymorphism studied has the same frequency distribution in both Raica and non-Raica people with or without diabetic conditions. The socio-economic and occupational environment of the members, which need to be explored further along with the genetic factors, may better account for the leanness and better control over the glucose levels in the Raica community.

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AUTHORS CONTRIBUTION

Dr. Mili Jain: Literature search, writing-original draft preparation, data collection, statistical analysis, and final approval of manuscript. Dr. Raj Kumar Vyas: Conceptualization, statistical analysis, writing-original draft and editing, final approval of the manuscript, and supervision.

CONFLICT OF INTEREST

None to declare.

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REFERENCES

1. World Health Organization. Diabetes. Available from: https://www.who.int/news-room/fact-sheets/detail/diabetes [Last accessed on 2023 Jun 13].
2. Ramachandran A, Snehalatha C, Kapur A, Vijay V, Mohan V, Das AK, *et al*. High prevalence of diabetes and impaired glucose tolerance in India: National Urban diabetes survey. Diabetologia 2001;44:1094-101. doi: 10.1007/s001250100627, PMID 11596662.
3. Ahuja MM. Epidemiological studies on diabetes mellitus in India. In: Epidemiology of Diabetes in Developing Countries. Vol. 1. New Delhi: Interprint; 1979. p. 29-38.
4. Ramachandran A. Epidemiology of type 2 diabetes in Indians. J Indian Med Assoc 2002;100:425-7. PMID 12674166
5. Mohan V, Pradeepa R. Epidemiology of diabetes in different regions of India. Health Adm 2009;22:1-18.
6. Hazarika CR, Babu BV. Prevalence of diabetes mellitus in Indian tribal population: A systematic review and meta-analysis. Ethn Health 2023;28:544-61. doi: 10.1080/13557858.2022.2067836, PMID 35469488
7. Kaul N, Ali S. Genes, genetics, and environment in type 2 diabetes: Implication in personalized medicine. DNA Cell Biol 2016;35:1-12. doi: 10.1089/dna.2015.2883, PMID 26495765
8. Hanis CL, Boerwinkle E, Chakraborty R, Ellsworth DL, Concannon P, Stirling B, *et al*. A genome-wide search for human non-insulin-dependent (type 2) diabetes genes reveals a major susceptibility locus on chromosome 2. Nat Genet 1996;13:161-6. doi: 10.1038/ng0696-161, PMID 8640221
9. Elbein SC, Hoffman MD, Teng K, Leppert MF, Hasstedt SJ. A genome-wide search for type 2 diabetes susceptibility genes in Utah Caucasians. Diabetes 1999;48:1175-82. doi: 10.2337/diabetes.48.5.1175, PMID 10331426
10. Cao H, Hegele RA. Human aryl hydrocarbon receptor nuclear translocator gene (ARNT) D/N511 polymorphism. J Hum Genet 2000;45:92-3. doi: 10.1007/s100380050018, PMID 10721670
11. Agrawal RP, Jain S, Shah S, Chopra A, Agarwal V. Effect of camel milk on glycemic control and insulin requirement in patients with type 1 diabetes: 2-years randomized controlled trial. Eur J Clin Nutr

2011;65:1048-52. doi: 10.1038/ejcn.2011.98, PMID 21629270

12. Hussain H, Wattoo FH, Wattoo MH, Gulfraz M, Masud T, Shah I, *et al*. Camel milk as an alternative treatment regimen for diabetes therapy. Food Sci Nutr 2021;9:1347-56. doi: 10.1002/fsn3.2078, PMID 33747450
13. Sboui A, Khorchani T, Djegham M, Agrebi A, Elhatmi H, Belhadj O. Anti-diabetic effect of camel milk in alloxan-induced diabetic dogs: A dose-response experiment. J Anim Physiol Anim Nutr (Berl) 2010;94:540-6. doi: 10.1111/j.1439-0396.2009.00941.x, PMID 19906135
14. Agrawal RP, Swami SC, Beniwal R, Kochar DK, Sahani MS, Tuteja FC, *et al*. Effect of camel milk on glycemic control, lipid profile and diabetes quality of life in type 1 diabetes: A randomised prospective controlled cross over study. Indian J Anim Sci 2003;73:1105-10.