MedCrave
Step into the World of Research
MOJ Anatomy & Physiology

Research Article

Association of fat patterning, hypertension and ACE (I/D) gene polymorphism: a study on two Tibeto-Burman linguistic group of Tripura, North East India

Volume 5 Issue 6 - 2018

Kusum Ghosh, Pranabesh Sarkar, Diptendu Chatterjee, Arup Ratan Bandyopadhyay
Department of Anthropology, University of Calcutta, India

Correspondence: Arup Ratan Bandyopadhyay, Department of Anthropology, University of Calcutta. 35, Ballygunge circular Road, Kolkata–700019, India, Email abamthro@caluniv.ac.in

Received: October 23, 2018 | **Published:** November 22, 2018

Abstract

Background: Hypertension is considered as a major cause of morbidity and mortality throughout the world and become a major global burden on public health in many developing countries. Regulation of blood pressure is a complex process. Apart from environmental factors, multiple genes have been responsible for hypertension. The gene-environment and fat patterning interaction in the pathogenesis of hypertension has not been extensively studied in the north-east Indian ethnic groups.

Aim: In this context, to best of our knowledge this is a maiden attempt to discern the association between fat patterning, blood pressure and ACE (I/D) gene polymorphism among the two Tibeto-Burman speaking ethnic groups (Chakmas and Tripuris) of North East India (Tripura).

Materials and methods: To achieve the purpose, total 293 (Chakma 148 and Tripuri 145) apparently healthy unrelated adult males from Tripura, North East India were incorporated in the present study. Anthropometric variables and physiological variables (blood pressure) have been collected using standard techniques. Extracted genomic DNA from mouthwash was PCR amplified and genotyped to understand ACE gene I/D polymorphism.

Results: Examination on the association of fat patterning and hypertension revealed significantly ($p<0.05$) higher hypertensive individuals and accumulation of central obesity among the Chakmas compared to the Tripuris. ACE (I/D) gene polymorphism demonstrated higher frequency of 'I' allele in the present study groups and the findings of the present study is in agreement with the distributions found in Asiatic populations and also in close corroboration with other Tibeto Burman linguistics groups of North East India.

Conclusion: Significantly ($p<0.05$) higher hypertensive individuals were found among the Chakmas in comparison to the Tripuris. The present study envisaged central adiposity is a major risk factor for hypertension in Chakmas rather than ACE (I/D) polymorphism.

Introduction

Non-Communicable Disease(s) (NCDs) includes cardiovascular disease, cancer, chronic lung diseases, and diabetes. Hypertension, overweight, physical inactivity, alcohol consumption are considered as the most important risk factors for NCDs (WHO, 2012). Most of the NCDs are multifactorial in nature. Both genes and environment are responsible for the prevalence of NCDs. Hypertension is often called "The silent killer" because it generally has no symptoms until serious complications develop.[1] Hypertension is considered as a major cause of morbidity and mortality throughout the world and become a major global burden on public health in many developing countries.[2] The prevalence of hypertension will be gradually increased up to 60% by 2025. In developed countries, the prevalence of hypertension ranges between 22% and 55%. In a recent report by World Health Statistics 2012 it was estimated that 63% of global death were occur due to non-communicable diseases (NCDs). The largest proportion of NCD deaths is caused by cardiovascular diseases (48%). The prevalence of hypertension for the European average was 44.2% compared with 27.6% in North America. In US the median prevalence of total hypertension in 2009 was 37.6% in men and 40.1% in women.[3] National Health and Nutrition Examination Survey (1999-2004) found that the prevalence of hypertension in the USA increased from approximately 27% to 29%. Similarly another survey by NHANES from 2004–2007 revealed a high prevalence of hypertension.[4]

The prevalence of hypertension varied among different studies in India, ranging from 2-15% in urban India and 2-8% in rural India. In developing countries, it is the seventh highest contributor to premature death. CVD was the largest cause of deaths in males (20.3%) as well as in females (16.9%) and it leads to 2 million deaths annually.[5] In India there is a trend of gradual increase in the prevalence of hypertension in last six decade. The prevalence of hypertension was found around 16.9% among Gujarat labor population.[6]

The Renin Angiotensin System (RAS) is an important system in regulating blood pressure and electrolyte balance. Genes encoding components of the RAS have been proposed as candidate genes that determine genetic predisposition to hypertension, diabetes and the risk of developing cardiovascular complications. Among the multiple genetic polymorphisms, the gene that possibly playing an active role in the pathogenesis of hypertension and cardiovascular disease is the angiotensin-converting enzyme (ACE) insertion/deletion (I/D) polymorphism. The ACE gene is mapped to chromosome 17q23 and is encoded by a 21 Kb gene that consists of 26 exons and 25 introns. Variations in the ACE gene, many of which influence levels of the protein, are associated with diseases such as hypertension, myocardial and One of the most important and widely studied variations in ACE is a 287-base pair (bp) Alu insertion/deletion (I/D) polymorphism in intron 16 (rs4340). The D allele was found to raise ACE plasma levels in Europeans.[7] The insertion/deletion (I/D) polymorphism of ACE is characterized by the presence (insertion) or absence (deletion) of a 28 bpAlu repeat sequence in intron 16 producing three genotypes (II homozygote, ID heterozygote and DD homozygote). Although the I/D polymorphism is located in a non-coding region (i.e. intron) of the ACE gene, several investigators have found that the D allele is related to increased activity of ACE in serum.[8] It is observed that the II genotype has lowest ACE levels while the DD type has the highest and finally the ID genotype has the intermediate level.[9]

India, occupying the Centre-stage of Paleolithic and Neolithic migrations, and somewhat has been under-represented in genome-wide studies of variation.[10] Being at the crossroads of migration, Indian populations have undergone complex and ancient admixture events over a long period[11–13] and have been the melting-pot of disparate ancestries originating from different parts of Eurasia and South-East Asia.[13] Although the date of entry of modern humans into India remains uncertain but it is reasonable to consider by the middle Paleolithic period (50,000–20,000 years before present [ybp]), humans appear to have spread onto many parts of India.[14] Contemporary ethnic India is a land of enormous genetic, cultural, and linguistic diversity.[15,16] A more recent study exploring Indian genomic diversity demonstrated four major ancestral genetic components in mainland India that included four dominant ancestries in populations from mainland India: Ancestral North-Indian (ANI), Ancestral South-Indian (ASI), Ancestral Tibeto–Burman (ATB) and Ancestral Austro-Asiatic (AAA).[13]

On the basis of the literature review on major and contributing publications regarding ACE I/D genetic polymorphism of the North East Indian population, it has been found that studies on the tribal population of Tripura seems to be under represented. In this backdrop, the present study, to best of the knowledge is the first attempt to understand the above mentioned issues on three Tibeto-Burman speaker endogamous ethnic populations (Chakmas - the migrant group and Tripuris–the aboriginal group) of North East India (Tripura).

Materials and methods

Present study was conducted among the one forty eight (148) participants of Chakmas, one hundred forty five (145) participants from Tripuri group. Prior to the study verbal consent were obtained from the participants.

Mouth wash was collected from all the participants in 15ml centrifuge tube. Genomic DNA was isolated from mouthwash following standard technique[17] with slight modification. PCR products were genotyped for I (Insertion) and D (Deletion) alleles of ACE gene using locus specific primer viz. Forward Primer-5'-CTG GAG ACC ACT CCC ATC CTT TCT-3' Reverse Primer-5'-GAT GTG GCC ATC ACA TTC GTC AGA T-3'. PCR product was taken for 2.5% agar gel electrophoresis containing ethidium bromide. The ACE I/D polymorphism consists of the presence (insertion, I) or absence (deletion, D) of 287-bp Alu repeat in intron 16 resulting in three genotypes: insertion homozygous (Ins/Ins), insertion-deletion (Ins/Del) heterozygous and deletion homozygous (Del/Del). Eventually, after electrophoresis, each DNA sample revealed one of the three possible patterns: a 597-bp band (Ins/Ins genotype), a 319-bp band (Del/Del genotype) or both 597-bp and 319-bp bands (Ins/Del genotype). Each sample found to have the Del/Del genotype was subjected to a second PCR amplification with insertion-specific primers to avoid possible Del/Del mistyping.[18] Blood pressure (Systolic blood pressures SBP and Diastolic blood pressure- DBP) were also taken following the standard technique[19] and classified in accordance with the Seventh Report of the Joint National Committee (JNC-7) recommendation.[20] The mean arterial pressure (MAP) was derived (Equation: MAP=[(2 x diastolic)+systolic]/3].[21] Allele frequencies were calculated by Maximum Likelihood Estimation.[22] Obtained data were doubly checked and analyzed using the SPSS (Version-16.0). Descriptive and inferential statistics were done in appropriate places and cut off was set as $p=0.05$.

The studied population

The Chakmas have inhabited the Chittagong Hill Tracts since long before the region became part of the Mughal Empire in 1666. Buddhism flourished in Bangladesh from the 5th to 12th centuries. Since the 13th century, Buddhism has been confined to the Chittagong area. The Chakma 'continued to acknowledge the suzerainty of the Arakanese king until 1784. According to the Census of India (2011) Chakmas in Tripura is 79813. Tripuris belong to linguistically within the Tibeto-Burman family. Tripuris are the largest tribal community in Tripura. They have first migrated in this territory and could be introduced as aboriginal tribe of Tripura. Numerically as per 2011 Census they are 592255 people in the State and Tripuris are numerically highest in number among all the tribal groups.[23]

Results

Anthropometric and physiological characteristics of the age matched studied populations (Table 1) demonstrated the Chakmas were significantly ($p<0.05$) taller than the Tripuris. Examination on Waist Circumference (WC) and Hip circumference (HC) revealed significantly ($p<0.05$) higher circumferences among the Chakmas in comparison to the Tripuris. However, BMI as general obesity marker did not show any difference between the two populations might be due to no difference in weight. Waist to Hip Ratio (WHR) as a measure of central obesity demonstrated significantly ($p<0.05$) higher central obesity among the Chakmas in comparison to the Tripuris. So far the Blood pressure as physiological variables concerns revealed significantly ($p<0.05$) higher SBP, DBP and derived MAP among the Chakmas compared to those of the Tripuris.

Table 1 Anthropometric and physiological characteristics of studied population

Variables	**Chakma (N=48) (Mean±SD)**	**Tripuri (N=45) (Mean±SD)**
Age (years)	36.07±12.65	35.62±12.03
Anthropometric		
Stature (cm)	165.53±4.88*	162.54±6.71
Weight (kg)	62.98±9.12	61.19±12.63
WC (cm)	87.18±10.128*	81.44±10.51
HC (cm)	92.70±9.32*	91.76±8.02
BMI	23.21±3.38	23.48±4.37
WHR	0.89±.08*	0.86±0.09
Physiological		
SBP (mm/Hg)	150.00±29.3*	133.72±19.2*
DBP (mm/Hg)	88.42±13.30*	81.22±13.20
MAP (mm/Hg)	108.94±16.65*	98.72±13.58

Distribution of genotypes of ACE (I/D) gene polymorphism (Table 2) demonstrated significant ($p<0.05$) difference in genotypes between the Chakmas and Tripuris and also revealed higher I alleles among the Chakmas in comparison to Tripuris.

Citation: Ghosh K, Sarkar P, Chatterjee D, et al. Association of fat patterning, hypertension and ACE (I/D) gene polymorphism: a study on two Tibeto-Burman linguistic group of Tripura, North East India. *MOJ Anat & Physiol.* 2018;5(6):368–371. DOI: 10.15406/mojap.2018.05.00227

Table 2 Distribution of genotype and allele frequencies of ACE (I/D) gene polymorphism of the studied populations

Population	Genotype			Allele frequency	
	II	ID	DD	I	D
Chakma	96 (64.86)	1 (0.68)	51 (34.46)	0.652	0.348
Tripuri	74 (51.03)	1 (0.69)	70 (48.28)	0.514	0.486

Figures in the (parenthesis) denotes the percentage

Distribution of hypertensive and normotensive groups classified in terms of SBP and DBP according to JNC 7 and ACE (I/D) genotypes including the allele frequencies among the studied populations presented in Table 3 & 4 revealed significantly ($p<0.05$) higher hypertensive individuals among the Chakmas compared to the Tripuris. Furthermore, the distribution of ACE (I/D) gene polymorphism demonstrated significant ($p<0.05$) between the two populations and higher I allele among the Chakmas in comparison to Tripuris for both the SBP and DBP.

Table 3 Distribution of Normotensive and Hypertensive (JNC7) according to SBP and the genotypes including allele frequency of ACE (I/D) gene polymorphism among the studied populations

Population	Hypertensive	Normotensive	Genotypes			Allele frequency	
			II	ID	DD	I	D
Chakma (n=148)	59(39.86)	89 (60.14)	96 (64.86)	1 (0.68)	51 (34.46)	0.652	0.348
Tripuri (n=145)	30 (20.69)	115 (79.31)	74 (51.03)	1(0.69)	70 (48.28)	0.514	0.486

Figures in the (parenthesis) denotes the percentage

Table 4 Distribution of Normotensive and Hypertensive (JNC7) according to DBP and the genotypes including allele frequency of ACE (I/D) gene polymorphism among the studied populations

Population	Hypertensive	Normotensive	Genotypes			Allele frequency	
			II	ID	DD	I	D
Chakma (n=148)	54 (33.78)	94 (66.28)	93 (64.86)	1 (0.68)	54 (34.46)	0.632	0.368
Tripuri (n=145)	36 (24.83)	109 (75.17)	80 (51.03)	1 (0.69)	75 (48.28)	0.556	0.444

Figures in the (parenthesis) denotes the percentage

Examination of blood pressure categories classified according to obesity pattern among the Chakmas (Table 5) and Tripuris (Table 6) revealed significant ($p<0.05$) differences in BMI (general obesity), WC and WHR (Central obesity) between hypertensive and normotensive individuals for both the populations.

Table 5 Distribution of Fat patterning according to hypertension and normotensive blood pressure among the Chakmas

Category		Hypertensive	Normotensive	P
BMI	Normal	18	90	0.05
	Overweight	36	1	
	Obese	2	1	
WC (cm)	Normal	17	65	0.05
	Risk	39	27	
WHR	Normal	15	85	0.05
	Risk	41	7	

Table 6 Distribution of Fat patterning according to hypertension and normotensive blood pressure among the Tripuris

Category		Hypertensive	Normotensive	P
BMI	Normal	11	86	0.05
	Overweight	9	35	
	Obese	3	1	
WC(cm)	Normal	42	92	0.05
	Risk	9	2	
WHR	Normal	46	83	0.05
	Risk	11	5	

Discussion

It would be apparent from the results, that the Chakmas vindicated significantly ($p<0.05$) higher hypertension and accumulation of central obesity than the Tripuris. The computed MAP (Table 1) indicated Hypertension Grade I[24] among the Chakmas compared to that of the Tripuris. The risk factors for NCDs including hypertension so far the central obesity measured through WC and WHR for Asian population.[25] The present study revealed significant ($p<0.05$) effect for Chakmas (Table 5) compared to the Tripuris (Table 6). Although there was no previous study regarding the ACE (I/D) gene polymorphisms of these two Tibeto Burman linguistic groups (Chakma and Tripuri), but the present study observed a higher frequency of 'I' allele in the present studied ethnic populations groups and the finding is in agreement with distributions found in Asiatic populations[26] and also in close corroboration with other Tibeto Burman linguistics groups like Jamatia and Mizo of North East India.[27,28]

Acknowledgements

Authors gratefully acknowledge the financial support from University of Calcutta [BI ((65) 8 & 9] and UGC- UPE 2 grant. Authors are thankful to the study participants and Dr. Rajesh Chatterjee of Tripura University for his untiring support to the fieldwork.

Conflict of interest

There is no conflict of interest whatsoever.

References

1. Siyad AR. Hypertension. *H J D Med.* 2011;3(1):1–16.
2. Foex P, Sear JW. Hypertension: Pathophysiology and treatment. *Continuing education in Anesthesia, Critical care and Pain.* 2004;4(3):71–75.

3. Olives C, Myerson R, Mokdad AH, et al. Prevalence, awareness, treatment, and control of hypertension in United States Countries, 2001-2009. *PLoSONE*. 2013;8(4):1–8.
4. Sarkar T, Singh NP. Epidemiological and genetics of hypertension. *J Assoc Physicians India*. 2015;63(9):61–68.
5. Kearney PM, Whelton M, Reynolds K, et al. Global burden of hypertension: analysis of worldwide data. *Lancet*. 2005;365(9455):217–223.
6. Tiwari RR. Hypertension and epidemiological factors among tribal labour population in Gujarat. *Indian J Public Health.* 2008;52(3):144–146.
7. Rigat B, Hubert C, Alhenc-Gelas F, et al. An insertion/deletion polymorphism in the angiotensin I-converting enzyme gene accounting for half the variance of serum enzyme levels. *J Clin Invest*. 1990;86(4):1343–1346.
8. Zarouk WA, Hussein IR, Esmaeil NN, et al. Association of angiotensin converting enzyme gene (I/D) polymorphism with hypertension and type-2 diabetes. *Bratisl Lek Listy.* 2012;113(1):14–18.
9. de Carvalho SS, Simões e Silva AC, Sabino Ade P, et al. Influence of ACE I/D polymorphism on circulating levels of plasminogen activator inhibitor 1, D-Dimer, ultrasensitive C-reactive protein and transforming growth factor β1 in patients undergoing hemodialysis. *PLoSONE*. 2016;11(3):e0150613.
10. Cann RL. Genetic clues to dispersal in human populations: retracing the past from the present. *Science*. 2001;291(5509):1742–1748.
11. Bamshad M, Kivisild T, Watkins WS, et al. Genetic evidence on the origins of Indian caste populations. *Genome Res*. 2001;11(6):994–1004.
12. Moorjani P, Thangaraj K, Patterson N, et al. Genetic evidence for recent population mixture in India. *Amer J Hum Genet*. 2013;93(3):422–438.
13. Basu A, Sarkar-Roy N, Majumder PP. Genomic reconstruction of the history of extant populations of India reveals five distinct ancestral components and a complex structure. *PNAS*. 2016;113(6):1594–1599.
14. Misra VN. Stone age in India: An ecological perspective. *Man & Env*. 1992;14:17–64.
15. Karve I. Hindu society: an interpretation. Poona, India: Deshmukhprakashan; 1961:171.
16. Majumder PP. People of India: Biological diversity and affinities. *Evolutionary Anthropology.* 1998;6(3):100–110.
17. Zayats T, Young LT, Mackey AD, et al. Quality of DNA extracted from mouth washes. *PLoS ONE*. 2009;4(7):e6165.
18. Shanmugam V, Sell KW, Saha BK. Mistyping ACE heterozygotes. *PCR Methods Appl.* 1993;3(2):120–121.
19. Lohman TG, Roche AF, Martorell R. *Anthropometric standardization reference manual*. Chicago: Human kinetics Books; 1988:1–12.
20. Chobanian AV, Bakris GL, Black HR, et al. The National High Blood Pressure Education Program Coordinating Committee Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. 2016.
21. Pocock G, Richards CD. *The human body: An introduction for the biomedical and health sciences*. 1st ed. New York: Oxford University Press Inc; 2009.
22. Cavalli-Sforza, LL, Bodmer WF. *The genetics of human populations*. San Francisco: WH Freeman and Company; 1971.
23. *http://www.tritripura.in/tri/Tribes/Index.aspx*
24. Kundu RN, Biswas S, Das M. Mean arterial pressure classification: a better tool for statistical interpretation of blood pressure related risk covariates. *Cardiology and Angiology: An International Journal.* 2017:6(1).
25. WHO Expert Consultation. Waist Circumference and Waist-Hip Ratio; 2008.
26. Zee RY, Lou YK, Griffiths LR, et al. Association of a polymorphism of the angiotensin I-converting enzyme gene with essential hypertension. *Biochem Biophy Res Commun.* 1992;184(1):9–15.
27. Farheen S, Basu A, Majumder PP. Haplotype variation in the ACE gene in global populations, with special reference to India, and an alternative model of evolution of haplotypes. *HUGO J*. 2011;5(1-4):35–45.
28. Borah PK, Shankarishan P, Ahmed G, et al. 2011 Polymorphism of angiotensin converting enzyme (insertion/deletion) and endothelial nitric oxide synthase (intron 4ab) genes in a population from northeast India. *J Genet*. 2011;90(3):e105–e109.

Citation: Ghosh K, Sarkar P, Chatterjee D, et al. Association of fat patterning, hypertension and ACE (I/D) gene polymorphism: a study on two Tibeto-Burman linguistic group of Tripura, North East India. *MOJ Anat & Physiol.* 2018;5(6):368–371. DOI: 10.15406/mojap.2018.05.00227