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Genetic Diversity among Coastal Populations of Maharashtra, Goa and Odisha, India

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ABSTRACT India has served as a gateway between continents, with waves of human migration leaving rich cultural and linguistic diversity in their wake. Studies of genetic variation provide inferences about the nature and intensity of forces that have modulated our evolutionary course. The researchers analyzed genetic diversity using 5 genetic markers (ABO, MN, Rh, Se and tasters/non-tasters) in seven populations sampled from Maharashtra, Goa (western coastal zone) and Odisha (eastern coastal zone) of India. The genetic distance estimated by the DISPAN program revealed that geographic segregation has led to genetic divergence among the population of similar ethnicity while population inhabiting same geographical region had higher genetic affinity despite their ethnic origin. ABO locus among all showed maximum genetic divergence between the populations. However, more studies referring to the coastal populations of India could provide new insights into the processes of admixture, selection and drift which lead to population dispersal and differentiation.

INTRODUCTION

India has served as major corridor for the dispersal of modern humans out of Africa ~2,000000 years ago (Cann 2001), owing to its geographical location at the tri-junction of the African, the northern Eurasian and the Oriental realm (Gadgil et al. 1997). The evolutionary antiquity of Indian ethnic groups and gene flow due to subsequent migrations has resulted in a rich tapestry of socio-cultural, linguistic and biological diversity of the subcontinent (Indian Genome Variation Consortium 2008). The tendency to isolate and subjugate the subordinate cultures has augmented such diversity, which is further nurtured by the ecological regimes variegation (Gadgil and Guha 1992).

Despite distinct religious communities, hierarchical castes and sub-castes and isolated tribal groups, inbreeding in human populations arising primarily from marriages between individuals related by ancestry or ethnicity have apparently hampered the gene flow between different factions resulting into isolated endogamous groups. Extensive literature has however demonstrated high degree of heterogeneity among Indian population (Cavalli-Sforza et al.1964; Papiha et al. 1996). The divergence in population genetic structure and sub-structure due to distinct genetic ancestry within or between racial/ethnic groups thereby results in population stratification. Furthermore, selective forces either favour the conservation of existing phenotypes or promote the emergence of new phenotypes leading to rapid divergence of traits between species and the depression of polymorphism within species (Vallender et al. 2004). Thereby, analyses of allele frequency of polymorphic loci such as ABO blood group, phenylthiocarbamide (PTC), taster, secretor etc. offer valuable inroads into understanding of phylogenetic relationships between the populations. Also, accuracy of evolutionary analysis of populations requires the analysis of a large number of genetic polymorphisms belonging to many loci (Bowcock et al. 1991).

Thus, the genetic diversity among people of India has long been of interest for understanding the origin and evolution of the people of the Indian subcontinent (Mountain et al. 1995; Majumder 1998; Basu et al. 2003; Cordaux et al. 2004), enabling us to understand the nature and intensity of actions of various forces that have modulated our evolutionary course. Despite numerous studies from different regions of India, the genetic variations among the coastal populations have not been studied much.

Objective of the Study

The present study was undertaken to assess the extent of genetic diversity and proximity in contemporary endogamous populations

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by estimating heterozygosity, genetic distance and genetic identity among the populations of Maharashtra, Goa from western coastal zone and Odisha from eastern coastal zone.

MATERIAL AND METHODS

Area and People

The present study includes data on the population groups from western and eastern coastal zones of India namely Maratha, Kunbi, Bhandari and Kharavi from district Ratnagiri, Maharashtra; Kharavi and Bhandari form South Goa district, Goa; and Nolia from district Ganjam, Odisha respectively. The data was collected in different phases during 2009-2011. Bhandari inhabiting western coastal region of Maharashtra and Goa derive their name from the Sanskrit word *'bhandar'* as in ancient times they were deployed as special guards for treasuries. Drawing toddy from palm tree to manufacturing liquor is their traditional economic resource. However post-independence, steadily growing population and disproportionate increase in palm tree have compelled them to do agriculture and other commercial pursuits. Bhandari included in the O.B.C category under Constitutional status, is an endogamous group following monogamy as a rule of marriage (Singh 2004).

Maratha is the dominant caste of Maharashtra mostly involved in agriculture. They produce food grains and other commodities for the consumption of the family and the surplus, if left is sold in the market. Those who do not own land works as agricultural labourers or industrial workers for wages. It is an endogamous group and speaks Marathi which belongs to Indo- European language group (Singh 2004).

The 'Kharavi' community of Maharashtra and Goa belong to Hindu religion despite the fact that after the Portuguese conquest of Goa in 1510 AD a large number of them were converted into Christianity. They reside along the coastal belt and are engaged in the traditional profession of constructing boats, their maintenance and fishing (Shaikh et al. 2011).

Nolias known as 'sea people' of Odisha sustain themselves by fishing and related activities. Except small children, every member in the household contributes in production activities. Adult men go into the sea, women and children help them on the beach and old people does mending of the fishing net and training young ones for fishing. They are Telugu fisherman who migrated to Ganjam district of Odisha in the end of nineteenth century and are now constitutionally categorised as OBC (Kapoor et al. 2010).

Kunbi in Maharashtra is a generic term which refers to individuals who depend on agriculture for their livelihood (Singh 2004). They belong to the Hindu religion and claim to have been warriors and land lord. They speak Marathi Language and make more than 30% population of Maharashtra (Tekade et al. 2011).

Data Collection

Study purpose was explained to all the volunteers prior to data collection and written consent was obtained from each subject. The study protocol was duly approved by the institutional ethical clearance committee. The blood samples were collected by finger prick with sterile lancet after cleaning the puncture site with 70% ethyl alcohol. ABO, Rh and MN blood groups typing of each individual was determined by standard agglutination method on glass slides using reagents Anti-A_1, Anti-B, Anti-D, Anti-M and Anti-N respectively. The secretor status was determined by using the haemagglutination inhibition test (Bhasin and Chahal 1996). Harris and Kalmus (1949) serial dilulation method was used to assess the taste sensitivity to PTC.

Data Analysis

Considering 4 alleles for ABO gene (O, A_1,A_2 and B) and 2 alleles for Rh gene (D, d), MN gene (M,N), Secretor gene (Se, se) and Taster gene (T,t), the allele frequencies were calculated for each population (Bhasin and Chahal 1996). The estimated allele frequencies of the genetic markers were further analysed to assess standard genetic distance (Nei 1972) and average heterozygosity (Nei 1978) using the genetic distance and phylogenetic analysis DISPAN program (Ota 1993). The parameter D measures the accumulated allele differences per locus or codon differences per unit length of DNA. The coefficient of genetic differentiation (G_{ST}) was used to estimate the level of gene flow or extent of gene diversity between the populations relative to the total population, expressed as

$G_{ST} = (D_{ST}/H_T)$

where, D_{ST} and H_T are the average gene diversity between populations and for total population respectively. The standard errors of average heterozygosity and Nei's genetic distance were computed for each population. The phylogenetic tree (dendrogram) was constructed by using the neighbor-joining (NJ) method (Saitou and Nei 1987) and the unweighted pair group-method with arithmetic mean (UPGMA) (Sneath and Sokal 1973) from distance matrices. Bootstrap resampling 1,000 (Efron 1982; Felsenstein 1985) was performed to test the percentage of a group's occurrence. Genetic Identity (I) was calculated (Hedrick 2005) as:

$$I = \frac{J_{xy}}{\sqrt{J_x J_y}} \text{ where } J_{xy} = \sum_{i=1}^{n} p_{i.x} p_{i.y}$$

$$J_x = \sum_{i=1}^{n} p_{i.x}^2 \text{ and } J_y = \sum_{i=1}^{n} p_{i.y}^2$$

Where, *Pi.x* and *Pi.y* are the frequencies of the 'i'th allele in two populations.

RESULTS

The phenotype frequency of genetic markers namely A_1A_2BO, Rh and MN blood groups, ABH secretion in saliva and taster trait were analysed to infer the allele frequency among the endogamous populations inhabiting Maharashtra, Goa and Odisha (Tables 1, 2 and 3). Generally, the frequency of O allele was highest followed by A_1, B and A_2 in all the populations under study. Incidence of O allele was higher among Maratha (0.66), A_1 among Bhandari (Maharashtra, 0.29), B allele among Nolias (0.27) and A_2 allele among Kunbi (0.15) as compared to the other populations. The frequency estimates of Rh alleles indicate higher value for Rh (D) than Rh(d) for all population, being highest among Kharavi of Maharashtra (0.86). Similarly, the allele frequency for secretor (Se) and taster allele (T) was higher for Bhandari (0.59) and Kunbi (0.60) of Maharashtra respectively.

Table 4 displays the average heterozygosity in the populations analysed for multiple loci. The average heterozygosity which indicates the degree of within-population variation was observed to be higher for Kunbi population (0.51) and lowest for the Maratha population (0.46).

Table 1: ABO blood group frequencies among different endogamous populations of Maharashtra, Goa and Odisha

Populations	*n*	*Phenotype*						*Allele frequencies*			
		A1	*A2*	*O*	*B*	*AIB*	*A2B*	*A1*	*A2*	*O*	*B*
Maharashtra											
Maratha	143	44(0.31)	4(0.03)	64(0.45)	21(0.15)	9(0.06)	1(0.01)	0.21	0.02	0.66	0.11
Kunbi	127	50(0.39)	1(0.01)	38(0.29)	26(0.21)	10(0.08)	2(0.02)	0.26	0.15	0.44	0.15
Bhandari	109	48(0.44)	1(0.01)	29(0.27)	24(0.22)	5(0.05)	2(0.02)	0.29	0.02	0.54	0.16
Kharavi	131	42(0.32)	5(0.04)	53(0.41)	18(0.14)	11(0.08)	2(0.02)	0.23	0.04	0.62	0.13
Goa											
Kharavi	137	39(0.29)	3(0.02)	58(0.42)	27(0.19)	9(0.07)	1(0.01)	0.19	0.02	0.64	0.15
Bhandari	137	56(0.41)	2(0.02)	39(0.29)	31(0.23)	8(0.06)	1(0.01)	0.27	0.02	0.55	0.16
Odisha											
Nolia	126	13(0.10)	3(0.02)	51(0.41)	46(0.37)	13(0.10)	0.00	0.11	0.01	0.61	0.27

Table 2: MN blood group and Rh gene frequencies among different endogamous population of Maharashtra, Goa and Odisha

Populations	*n*	*Phenotype*			*Allele frequencies*		*Phenotype*		*Allele frequencies*	
		M	*N*	*MN*	*M*	*n*	*Rh+(D)*	*Rh-(d)*	*D*	*d*
Maharastra										
Maratha	143	41(0.31)	14(0.11)	78(0.59)	0.60	0.39	137(0.79)	6(0.21)	0.79	0.21
Kunbi	127	55(0.43)	43(0.34)	29(0.23)	0.55	0.45	119(0.75)	8(0.25)	0.75	0.25
Bhandari	109	48(0.44)	28(0.26)	33(0.30)	0.59	0.41	103(0.77)	6(0.24)	0.77	0.24
Kharavi	131	58(0.44)	21(0.16)	52(0.40)	0.64	0.36	127(0.83)	4(0.18)	0.83	0.18
Goa										
Kharavi	137	60(0.44)	31(0.23)	46(0.34)	0.61	0.39	131(0.79)	6(0.21)	0.79	0.21
Bhandari	137	61(0.45)	38(0.28)	38(0.28)	0.58	0.42	129(0.76)	8(0.24)	0.76	0.24
Odisha										
Nolia	122	55(0.40)	15(0.11)	67(0.49)	0.65	0.35	113(0.73)	9(0.27)	0.73	0.27

Table 3: Secretor and taster gene frequencies among different endogamous population of Maharashtra, Goa and Odisha

Population	*n*	*Phenotype*		*Allele frequencies*		*n*	*Phenotype*		*Allele frequencies*	
		Secretor (Se)	*Non-secretor (se)*	*Se*	*se*		*Tasters (T)*	*Non-tasters(t)*	*T*	*t*
Maharashtra										
Maratha	143	125(0.87)	18(0.13)	0.65	0.36	174	128(0.74)	46(0.26)	0.49	0.51
Kunbi	127	103(0.81)	24(0.19)	0.57	0.44	127	107(0.84)	20(0.16)	0.60	0.39
Bhandari	109	91(0.84)	18(0.17)	0.59	0.41	109	86(0.79)	23(0.21)	0.54	0.46
Kharavi	131	103(0.79)	28(0.21)	0.54	0.46	131	92(0.70)	39(0.29)	0.45	0.55
Goa										
Kharavi	137	114(0.83)	23(0.17)	0.59	0.41	125	98(0.72)	27(0.20)	0.56	0.44
Bhandari	137	108(0.79)	29(0.21)	0.54	0.46	130	104(0.76)	26(0.19)	0.56	0.44
Odisha										
Nolia	126	114(0.91)	12(0.09)	0.69	0.31	120	76(0.63)	44(0.37)	0.39	0.61

Table 4: Average heterozygosity and its standard error for all loci in populations of Maharashtra, Goa and Odisha

Population	*Heterozygosity*	*SE*
Maharashtra		
Maratha	0.456	0.034
Kunbi	0.509	0.052
Bhandari	0.487	0.039
Kharavi	0.461	0.045
Goa		
Kharvi	0.464	0.034
Bhandari	0.489	0.036
Odisha		
Nolia	0.462	0.025

Table 5: Gene diversity analysis for individual loci in populations of Maharashtra, Goa and Odisha

Locus	D_{ST}	H_T	H_S	G_{ST}
A1A2BO	0.01249	0.588	0.576	0.021
MN	0.00002	0.004	0.477	0.004
Rh	0.00003	0.005	0.349	0.005
ABH	0.00540	0.482	0.477	0.011
Taster	0.00033	0.018	0.491	0.018
All	0.00615	0.480	0.474	0.013

The extent of gene diversity as expressed by heterozygosity indices (H_S, H_T, D_{ST}) and coefficient of gene differentiation (G_{ST}) have been furnished in Table 5. The value for H_S showed within population genetic variability to be highest for ABO locus (0.576) and lowest for Rh locus (0.349). Between populations gene diversity was also higher for ABO locus (0.0125) but lowest for MN locus (0.00002). By comparison, the measure of genetic differentiation (G_{ST}) revealed maximum genetic divergence between population relative to the combined population for ABO locus (0.021) followed by taster (0.018) and secretor trait (0.011) though the differentiation was not so pronounced.

In Maharashtra (Table 6), the distance was found to be higher between Kunbi and Kharavi (1.94%) followed by Maratha (1.88%) however, the latter seemed to be more closely related to Bhandari and Kharavi (0.0029). Kharavi and Bhandari from Goa suggested higher genetic affinity (0.00008). While geographic differentiation has lead to divergence in the population belonging to similar ethnic background that is,

Table 6: Genetic Distance Matrix along with standard error among different populations of Maharashtra, Goa and Odisha

	Maharashtra				*Goa*	
	Maratha	*Kunbi*	*Bhandari*	*Kharavi*	*Kharavi*	*Bhandari*
Maharashtra						
Kunbi	0.0188±0.0117					
Bhandari	0.0029±0.0034	0.0043±0.0051				
Kharavi	0.0029±0.0038	0.0194±0.0096	0.0044±0.0019			
Goa						
Kharavi	0.0001±0.0018	0.0114±0.0113	0.0003±0.0033	0.0030±0.0033		
Bhandari	0.0070±0.0040	0.0041±0.0060	-0.0025±0.0011	0.0054±0.0039	0.0008±0.0023	
Odisha						
Nolia	0.0093±0.0052	0.0394±0.0189	0.0186±0.0095	0.0163±0.0081	0.0164±0.0084	0.0257±0.0112

Table 7: Genetic identity matrix among different populations of Maharashtra, Goa and Odisha

	Maharashtra				*Goa*	
	Maratha	*Kunbi*	*Bhandari*	*Kharavi*	*Kharavi*	*Bhandari*
Maharashtra						
Kunbi	0.9812					
Bhandari	0.9971	0.9957				
Kharavi	0.9971	0.9806	0.9956			
Goa						
Kharavi	0.9999	0.9886	0.9997	0.9970		
Bhandari	0.9930	0.9959	1.0025	0.9946	0.9992	
Odisha						
Nolia	0.9907	0.9606	0.9814	0.9837	0.9836	0.9743

Kharavi (0.0030) and Bhandari (0.0025) from Maharashtra were at greater distance from their respective stock inhabiting Goa. The genetic identity matrix presented in Table 7 also confers to the analysis of genetic proximity.

The dendrogram drawn on the basis of genetic distance matrix as per UPGMA method, consists of three major clades constituted by Nolia, Kunbi and Bhandari (Goa), Kharavi (Goa) Bhandari (Maharashtra), Kharavi (Maharashtra) and Maratha (Fig. 1). Nolias demonstrated earliest divergence followed by Kunbi as compared to the other populations. Clade comprised of Bhandari (Goa), Kharavi (Goa), Bhandari (Maharashtra), Kharavi (Maharashtra) and Maratha depicts greater affinity to each other. Within this broad cluster there are two sub-clusters: the first formed by Bhandari (Goa) and Bhandari (Maharashtra); and the second by Kharavi (Goa) and Maratha.

DISCUSSION

The multiple waves of migration from time immemorial have supplemented the contemporary ethnically, culturally, linguistically and genetically diverse populations. Despite the anthropological and archaeological evidences contentious issues about the origin and divergence of populations in the Indian subcontinent remains inexplicable. Reddy et al. (1987) showed that anthropometric pattern of variation documents the importance of geographic proximity of micro-differentiation under the Indian social setup. Genetic measures inclusive of genetic distance, heterozygosity and genetic differentiation permit analysis of divergence and affinity among populations.

In the present study, the average heterozygosity for all the population was high ranging from 0.456 for Maratha to 0.509 for Kunbi. The low gene differentiation 0.004 to 0.021 for MN

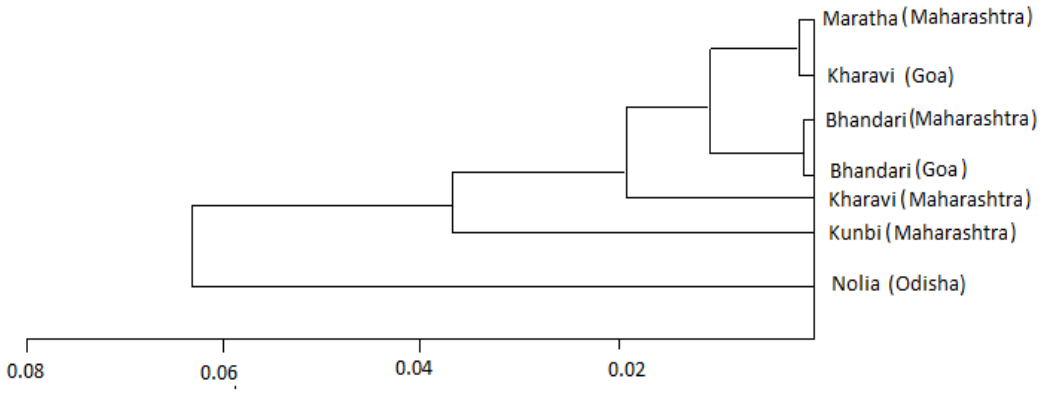

Fig. 1. Dendrogram matrix among coastal population of Western and Eastern coastal zones

and A1A2BO locus respectively concord with the finding of low (0.26-1.70%) genetic differentiation in Indian populations by Papiha et al. (1982) asserted on the basis of analysis of genetic markers consisting of 11 blood groups and red cell enzyme systems in 14 endogamous groups of north-west India. Mukherjee et al. (1979) analysed 16 biomarker among nine endogamous population of Maharashtra (Nava Budha, Maratha, Deshastha Rigvedi Brahmin, Chitpavan Brahmin, Chandrasenya Kayastha Prabhu, Parsis, Bhil, Pawara and Katkari). The genetic divergence between these groups was small as compared to the within group heterogeneity and the average heterozygosity per gene per locus ranged 20–22%. In the present study, intra population gene diversity H_S were quite high compared to inter-population gene diversity. Previous studies on Indian population have also revealed higher intra-population diversity (Bamshad et al. 1998; Tekade et al. 2011). This suggests the differences in genetic structure are more likely to be due to their breeding structure, differential migration and ethnic affiliation.

It has been observed that many genetic studies have been carried on the different populations representing different part of India and the genetic composition of the Indian people has been examined according to the existing social structure. The coastal populations or fishing communities of Western coastal zone and Eastern coastal zone have not been studied so far. Such studies can throw light on the various process of admixture, selection and drift which lead to population dispersal and differentiation and might be of special interest to cultural biologist, anthropologists, historians, human genetist and geographers.

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