

Diversity Among Selected Caste and Tribal Groups of Western India: Origin, HLA and Disease Associations

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ABSTRACT Indian population is well known for its genetic diversity. Among the numerous endogamous communities, which is restricted very much by custom, marriage and occupation, a compiled analysis of 1742 individuals comprising of different caste and tribes from Maharashtra Western India is presented. The sympatrically isolated caste and tribal population of western India with different origins, migration pattern and breeding habits differed significantly in their HLA and also in their HLA allele (Relative risk) prevalence for a number of disease associations. Further the analysis reveal that the caste groups of India cannot be considered as a single panmictic population with reference to genetic characteristics, which may have a clinical relevance in unrelated donor selection for allogenic Bone marrow transplantation and disease associations in India.

INTRODUCTION

The caste system in India (Bhasin and Walter 2001) is characterized by class exogamy and caste endogamy and in biological terms each caste/tribe is a breeding isolate. Having differed in origin, migration, linguistics, socio-cultural aspects and settlement these populations diverged further. Whether such sympatric breeding isolates would be differentially susceptible to a given epidemic in a given region/city, or be subject to similar evolutionary pressures, requires an understanding of evolutionary principles. In drawing lessons from community genetics, one has to remember that the gene pool, linguistics and culture can be regarded as three independent entities which may overlap in terms of time, space, origin and spread, but not as a rule. By virtue of their immune repertoire, the diverse gene pools of India may not be equally susceptible to an epidemic of infection. Different genes may be involved in different population (Hill 1998).

THE HLA SYSTEM

The HLA system, most polymorphic and complex set of genetic markers known in man, is of valuable significance in anthropological studies (Bhasin et al. 1994). These cell-surface antigens play an important role in the activation of immune competent cells, thereby promoting

immune response. The extensive polymorphism of the HLA system is associated with a large peptide repertoire for initiating immune responses against a wide range of foreign antigens (pathogens). They also have useful application in the study of the origin, evolution and migration patterns of human populations. Distribution of HLA antigens in various population groups of the India has been reported (Pitchappan et al. 1984; Rajasekar et al. 1987; Balakrishnan et al. 1996; Agrawal et al. 1999; Shankarkumar et al. 1999; Chayya et al. 2000; Jaini et al. 2002). For the indigenous caste/tribal populations of India practically very little information on the distribution of HLA antigens exists. The present study is the first population genetic analyses of HLA antigens in endogamous caste and tribal groups of Western India. Population specific distribution of HLA alleles is necessary and interesting both in population genetics and in HLA disease association studies (Bodmer 1987). Large number of this kind of studies have other uses e.g. Likelihood of finding an unrelated HLA compatible stem cell donor for allogenic stem cell transplantation and also for constructing a National stem cell registry in India. The Indian population differs in their origin, migration and settlement, which correlate to the advent of social stratification and caste system which, dates back to the existence of human societies in the Eurasian steppes. Analysis of genetic data

suggests that the inhabitants of the Western part of the Eurasian steppes, originally settled by Caucasoid people speaking Indo-European languages, migrated in various directions, including Iran and India. It is believed that these pastoral nomads often generated hierarchical societies and introduced the caste system in Indian subcontinent (Cavali-Sforza et al. 1993).

Western Indian Community

HLA distribution from 1742 healthy individuals belonging to different caste and tribal groups from Maharashtra was compiled in Table 1 and Table 2. The origin and caste/tribe details of these populations reported are explained in detail elsewhere (Einthoven 1990). The HLA alleles associated among these population groups with various diseases have been presented in Table 3.

HLA Typing

Ten to fifteen milliliters of venous blood (in heparin 50 IU/ml) was collected in a sterile tube from each individual after obtaining ethical consent. The lymphocytes were isolated by density gradient centrifugation on Histopaque (Boyum 1968). HLA A, B, C and DR locus antigens were identified by NIH two – stage Microlymphocytotoxicity assay (Terasaki and McClelland 1964) using T cells for class I typing and B cells isolated by a miniature nylon wool column for class II with a longer incubation period (Manikasundari et al. 1984). A total of 190 indigenous (Shankarkumar et al 1998) as well as commercially procured antisera were used for defining 17 specificities for HLA A locus, 29 for HLA B locus, 8 for HLA C locus and 10 for HLA DR locus antigens. The typing tray included a minimum of three antisera for each supertypic specificity. Further the molecular typing of HLA alleles were done using the extracted genomic DNA and the Polymerase chain reaction – sequence specific priming (PCR-SSP) kits.

Statistical Analysis

The phenotype frequencies, gene frequencies, relative risks (Shankarkumar et al. 2002) presented in Tables 1, 2 and 3 were estimated using our database and computer programs. Since each individual is tested for several HLA alleles and the same data used for comparing the

frequency; it is possible that one of the alleles will by chance deviate significantly. To overcome this error, the P value is corrected by use of Bonferoni inequality method (Dunn 1961) i.e. by multiplying it with the number of alleles compared and the quality of the study design was assessed from the published data.

Theoretically high polymorphism of a gene can occur due to mutation rate, selection, genetic hitchhiking or a combination of all the three (Kaufman 1996). Indigenous populations or caste/tribal groups show a very restricted diversity of alleles at a particular HLA loci consistent within a population. Moreover specific alleles found uniquely in a particular indigenous group as HLA B48 among Patels caste, B14 among Parsis and Badaga tribes, B21 among Koya tribe etc. may have been generated by point mutation or gene conversion from the ancestral allele after the group separated from the other groups (Shankarkumar et al. 2002). Multiple polymorphic alleles in a population are maintained at appreciable frequencies due to either overdominance (heterozygous advantage), frequency dependent selection, bottleneck effect or other selective forces (Shankarkumar et al. 2002). Both selective forces and a high rate of germline diversification are involved in the evolution of HLA allelic diversity.

Immunological Diversity: Are They Evolutionary?

The immune system is the most important biological system affected by the evolutionary pressures of diseases and is characterized by very high levels of polymorphism. The implications of these polymorphic diversities are important in community genetics in general and western India in particular, given the variation in HLA allele repertoires of various western Indian caste (Table 1) and tribal (Table 2) populations. The tables show the frequencies of various alleles in different regions of Western India, stratified data based on the exact caste and tribal groups revealed greater diversity and genetic distances equivalent to two globally distant populations (Shankarkumar et al. 1999; Shankarkumar et al. 2000; Chayya and Shankarkumar 2001). The numerical strength and spatial distribution of various exogamous clan of a caste/tribe decides the marriageable range which defines the gene pool. Our study on the Brahmins from Maharashtra

Table 1: Percentage frequencies among different Western Indian caste groups

HLA Ref	Banya 15 N=965	Lohana 15 N=46	Brahmin 18 N=54	Maratha 20 N=289	Jains 19 N=161	CKP 15 N=50	Kunbi 18 N=26	MaharV.Prajpathi 18 N=32	Parsee 15 N=50	Patels 15 N=67	Patels 15 N=112
A1	24.4	21.7	46.3	28.37	32.3	16	15.4	25	16	22.4	45.5
A2	35.9	23.9	16.7	30.8	22.4	30	53.8	40.6	30	9	31.3
A3	21.8	26.1	22.2	9	19.3	20	3.8	9.4	20	4.5	22.3
A9	33.3	28.3	24.1	25.26	36	64	50	28.1	64	67.2	40.2
A10	9	23.9	11.1	8.3	13.7	0	11.5	6.3	0	14.9	7.1
A11	20.5	19.6	24.1	24.57	24.2	10	11.5	18.8	10	14.9	33
A19	10.3	26.1	14.8	21.11	15.5	56	42.3	53.1	56	62.7	10.7
A28	12.8	13	9.3	12.8	16.1	0	7.7	12.5	0	0	0
B5	41	23.9	38.9	17.3	36	54	15.4	12.5	54	29.9	28.6
B7	7.7	13	22.2	23.88	4.3	26	23.1	28.1	26	3	13.4
B8	10.3	10.9	3.7	6.23	9.3	0	7.7	6.3	0	4.5	8
B12	17.9	21.7	16.7	10.03	13.7	6	15.4	15.6	6	26.9	17.9
B13	1.3	13	1.9	5.88	6.2	10	3.8	6.3	10	28.4	1.8
B14	0	4.3	1.9	0	0.6	0	0	0	0	41.8	3.6
B15	10.3	4.3	9.3	5.19	9.3	22	23.1	9.4	22	7.5	33
B16	1.3	0	0	0	1.2	0	0	0	0	0	0
B17	17.9	30.4	18.5	12.8	15.5	0	3.8	21.9	0	9	11.6
B18	3.8	8.7	5.6	2.42	9.3	0	0	3.1	0	0	8
B21	10.3	6.5	7.4	4.5	5.6	0	0	0	0	0	12.5
B22	5.1	4.3	14.8	3.81	4.3	6	7.7	3.1	6	6	11.6
B27	3.8	4.3	1.9	11.76	3.7	10	11.5	0	10	0	2.7
B35	21.8	19.6	14.8	26.3	25.5	12	30.8	25	12	23.9	25.9
B37	0	2.2	1.9	5.19	1.2	6	3.8	0	6	1.5	0
B40	29.5	13	20.4	20.42	26.1	22	38.5	43.8	22	13.4	17
B48	0	0	0	0	0	0	0	0	0	0	0.9
Cw1	2.6	2.2	3.7	7.6	0	*	*	*	*	*	8.7
Cw2	2.6	6.5	0	4.3	2.5	*	*	*	*	*	8.7
Cw3	10.3	19.6	7.4	29.3	14.3	*	*	*	*	*	13
Cw4	30.8	34.8	25.9	19.6	31.7	*	*	*	*	*	82.6
Cw5	0	6.5	7.4	1.1	6.2	*	*	*	*	*	4.3
Cw6	3.8	4.3	3.7	20.7	2.5	*	*	*	*	*	39.1
Cw7	2.6	0	1.9	3.3	5	*	*	*	*	*	0
DR1	0	0	10.3	16.84	5.6	*	*	*	*	*	*
DR2	51.4	22.2	37.9	50.53	33.8	*	*	*	*	*	*
DR3	32.4	27.8	31	7.37	22.5	*	*	*	*	*	*
DR4	21.6	11.1	24.1	8.42	33.8	*	*	*	*	*	*
DR5	27	27.8	27.6	15.79	35.2	*	*	*	*	*	*
DR6	5.4	0	6.9	12.63	0	*	*	*	*	*	*
DR7	29.7	44.4	24.1	29.47	35.2	*	*	*	*	*	*
DR8	0	0	0	70	0	*	*	*	*	*	*
DR9	0	0	0	7.37	1.4	*	*	*	*	*	*
DR10	0	0	0	14.74	0	*	*	*	*	*	*

(Shankarkumar et al. 2002) and Maratha caste (Shankarkumar et al. 2001) revealed that they differ in their allele and haplotype frequencies. This indicates that various caste/tribal population differing in their origins, migration and settlement adopted a belief, like Hinduism and identified themselves with a pre-existing or a new professional group such as Marathas, Bhils, Pawras, Mahars, Kunbis, Jains, Bhramins, Patels, or Parsis. Our recent DNA based HLA class I studies has further confirmed their remarkable level of allelic diversity (Shankarkumar et al. 2002; Shankarkumar et al. 2002).

Differential Disease Susceptibility: Caste System?

There are few studies that describe the varied HLA disease susceptibility from populations living in the same area. We have compiled the different diseases which have revealed significant HLA associations (Table 3) among western Indians. Further we have reported new novel allele associations in the Western Indian populations (Shankarkumar et al. 2002). We have investigated the association of HLA B27 in Ankylosing spondylitis (Shankarkumar et al.

Table 2: Percentage gene frequencies among different Indian tribals

HLA	Orans N-48	Pawra N-38	Bhils N-53	Malayalis N-42	Irulas N-191	Koya N-94	Kota N-103	Badagas N-58
Ref	Present	17	17	17	17	17	17	17
A1	9.9	9.7	16.4	4.9	6.8	3.7	7.77	9.48
A2	8.7	33.1	24.8	18.4	10.8	23.4	45.68	17.24
A3	11	8.2	7.9	1.2	5.1	5.3	6.8	13.79
A9	48	14.2	16.4	19.8	22.1	13.3	5.34	8.62
A10	0	6.8	8.9	4	10.5	4.8	2.91	5.17
A11	12.2	14.2	9.9	22.8	18.6	28.7	16.05	13.79
A19	20.9	18.9	19.9	15.1	11.9	0.5	2.91	12.07
A28	1	5.4	3.8	8.7	2.9	1.1	0.49	5.17
B5	13.4	4	11	11.4	23.8	4.8	7.28	6.9
B7	9.9	14.2	9.9	11.4	1.6	8	15.63	3.45
B8	0	5.4	8.9	2.4	14.1	4.3	1.46	4.31
B12	25	4	2.9	3.6	1.1	10.1	3.4	11.21
B13	7.6	2.7	0.9	7.4	5.1	3.2	2.48	7.76
B14	0	0	0	0	0.3	1.6	2.91	3.45
B15	9.9	1.3	4.8	11.4	1.1	1.1	3.88	8.62
B16	0	*	*	*	*	1.1	0	0
B17	4.3	8.2	8.9	11.4	8.8	25.5	2.43	3.45
B18	1	0	0	*	*	2.2	0.49	1.72
B21	0	*	*	*	*	0.5	0	0
B22	0	2.7	2.9	15.5	2.9	0.5	0.49	9.48
B27	2.1	1.3	3.8	*	*	1.6	1.91	4.31
B35	20.9	17.3	11	1	9.9	2.7	3.4	6.03
B37	1	4	1.9	0	1.3	0	*	*
B40	13.4	27.5	22.3	15.5	18.5	5.3	11.17	12.07
B53	0	15.7	14.2	0	4	1.1	*	*

* Not reported/ done

All values are percentages. Highest/ higher frequencies of specific allele are indicated in Bold.

Table 3: HLA and disease associations observed in western Indian population

Disease	Associated HLA	Relative Risk	Reference Number
Leprosy	B40	3.14	3
Leprosy	A*1102	30	37
Multiple Sclerosis (MS)	B12	*	39
Multiple Sclerosis (MS)	A11,B16,Cw7	2.6,13.8,5.46	17
Multiple Sclerosis (MS)	DRB1*15	16.15	17
Lymphoid Leukaemia	B35	*	20
Rh(D) isoimmunization	A3,B17,Cw2,DR4	2.6	35
Psoriasis	A1,B17,Cw6,	2.76,3.11,2.98,	11
Psoriasis	DR7,DQw3	2.15,3.16	11
Psoriasis	C4BQ0	10.73	11
Ankylosing Spondylitis (AS)	B27	273	4
Ankylosing Spondylitis (AS)	B27	71.5	16
Ankylosing Spondylitis (AS)	B27	72.22	26
Haemophilia with Synovitis	B27	34.6	34
Systemic Lupus Erythematosus (SLE)	DRB*03,DQB*0302	9.67,8.02	36

* Not reported.

All associations were valid in the total Western Indian populations

Values in Bold are highest Relative risks in a population

2002) and a significantly stronger association with HLA B*2714 was identified among the kunbi caste. The kunbi are landholders and husbandman who earn their living as field laborers, as

carriers, as gardeners or house servants. They are mainly distributed among Maharashtra State. The association can be attributed to the genetic phenomenon of hitchhiking i.e. the disease spread

with the migration of the community and the causative genes are linked to the HLA allele. In a similar manner HLA B12 association was observed with multiple sclerosis among Parsis (Trikannad et al. 1982) and other western Indians (Bale et al. 1980, 1982; Mehta et al. 1987; Chablani et al. 1992; Kankonkar et al. 1998; Shankarkumar et al. 2002a, b, 2003; Kankonkar et al. 2003). Thus the origin, migration, sympatric isolation and the resultant divergence of the gene pool at HLA loci add a new dimension to community genetics in terms of disease and prevalence.

DISCUSSION

The present analysis reveal, the heterogeneous nature of the Indian population suggesting that the population as such or even a linguistic or regional population within it cannot be considered as a panmictic pool; only a caste group may be considered as a homogenous gene pool with its diverse haplotype combinations and high rates of consanguinity. However, it is not known how these HLA allele dependant selection mechanisms might have influenced the disease pathogenesis among the Western Indian population groups. Further studies on non-recombinant Y chromosome (NRY) and mitochondria DNA (mt DNA) alleles can be used to trace migration pattern of different caste and tribal groups of western India.

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