

HLA Antigen Distribution in Maratha Community from Mumbai, Maharashtra, India

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ABSTRACT Two hundred and eighty nine unrelated Marathas residing in Mumbai, Maharashtra, (Western India) were studied for HLA A, B, C and DR locus antigen profiles. The HLA antigen maximum likelihood gene frequencies of HLA A1, A2, A9 (24), A11, A19 (33), B5, B7, B35, B40 (61), Cw3, Cw6, DR2, DR5, DR7, DQ1 and DQ2 were increased while that of HLA A3, A10 (26), A36, B8, B13, B16 (38), B18, B21, B22 (55), B53, B73, Cw5, Cw7, DR3, DR4, DR9, and DQ3 were decreased in the Marathas. HLA antigens A25 (10), B14, B39 (16), B54 (22), B56 (22), B58 (17) and Cw8 were not identified in the present investigation. Two Locus haplotype analyses revealed the presence of A10-B8, A1-B17, A24-B52, B5-Cw9, B13-Cw3, B15-Cw2, B35-Cw4, DR2-DQ1, DR5-DQ3 and DR1-DQ9 haplotypes with positive linkage disequilibrium among the marathas. Haplotype A2-B12 was the only haplotype identified in negative linkage disequilibrium. The observed maximum likelihood gene frequencies, haplotype frequencies and linkage disequilibrium in Marathas suggest the influence of genetic drift caused by selection, geography and culture. Further the study reveals that the Hindu population of India cannot be considered as a single panmictic population due to vast allelic diversity and immense heterozygosity in haplotypes.

INTRODUCTION

India is thought to have been one the site of earliest human settlements. Subsequently the region has been subjected to successive waves of immigration and invasions from the Middle East, Central Asia and Mongolia, contributing to the present day gene pool (Bhasin et al. 1994). The population exhibits not only a wide variety of ethnic but also great cultural and linguistic diversity. Numerous endogamous ethnic groups delineate within each linguistic or religious group based on biological and socio-cultural characteristics. Maharashtra, the Western state of India, lying between 74° - 78° E longitude and 18° - 20° N latitude with Mumbai as its capital city consists of 32 districts. Hindus form 81.94% of the population while others are Muslims (8.4%), Christians (1.42%), Buddhists (6.48%), Sikhs (0.2%) and Jains (1.64%) (Dikshit 1986).

A caste is a group of people having a specific

social name defined generally by descent, marriage and occupation. Each caste has its own customs that restrict the occupation and dietary habits of its members and their social contact with members of other castes. There are about 3000 castes, and more than 25,000 sub-castes in India, numbering from a few hundreds to few millions. According to the traditional law books castes are grouped into four Varnas: in hierarchy order, viz.: first the Brahmins (priests and scholars), then the Kshatryas (warriors and rulers), then the Vaishyas (merchants, traders and farmers) and lastly the Sudras (artisans, laborers, servants and slaves). Caste members marry only members of their own caste (endogamy). The Maratha caste comprises about 40 million Marathi speaking people living mainly in Maharashtra, Western India. They are mostly farmers, who are believed to have originated from the region that extends from Mumbai to Goa. They are further subdivided into 96 Kuli Marathas based on their surnames. In the 17th and 18th century they formed a powerful military confederacy in rivalry with the Mogul Emperors. The HLA system, most polymorphic and complex set of genetic markers known in man, is of valuable significance in anthropological studies (Bhatia and Rao 1986; Dausset 1981). Distribution of HLA antigens in various ethnic groups of the world have been reported (Mehra et al. 1998). Few reports are available on immigrant Indian population (Imanishi et al. 1992; Singal 1972) and regional Hindu populations (Mittal et al. 1982; Raha 1975; Pitchappan et al. 1984; Mehra et al. 1986; Selvakumar et al. 1988; Rajalingam et al. 1997; Mehra et al. 1994; Munkhbhat et al. 1997; Agrawal et al. 1999; Shankarkumar et al. 1999). However data is rather scanty from western part of India (Papiha 1996). Therefore we present here the immunogenetic profile from Maratha caste group living in Mumbai.

MATERIALS AND METHODS

Blood samples from random 289 healthy un-

related Marathas residing in Mumbai were studied for HLA -A, -B, -C and -DR Loci antigens. Their population specific details and genealogy were recorded in the precoded questionnaire. The average mean age among the samples were 30 ± 2 - 40 ± 2 . The ratio between the males and females were 3:4. Five to ten milliliters of venous blood (in heparin 50 IU/ml) was collected in a sterile tube from each individual. 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 longer incubation period (Manikasundari et al 1984). A total of 190 anti HLA 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 antisera were commercial (Biotest, Germany; Behring, Germany; Pelfreez, USA) as well as indigenous (Shankarkumar et al 1998) in origin. The typing tray included a minimum of three antisera for each supertypic specificity. The phenotype frequency (PF), gene frequency (GF), standard error of gene frequency (SEGF), haplotype frequency (HF), Co-efficient of linkage disequilibrium (Delta) and t values were calculated following the methods described by Baur and Daniloves (1980).

RESULTS

The results on HLA A, B, C and DR antigen maximum likelihood gene frequencies and standard error of gene frequencies in 289 unrelated healthy Marathas are presented in table 1. The frequencies of HLA A1, A2, A9 (24), A11, A19 (33), B5, B7, B35, B40 (61), Cw3, Cw6, DR2, DR5, DR7, DQ1 and DQ2 were increased while HLA A3, A10 (26), A36, B8, B13, B16 (38), B18, B21, B22 (55), B53, B73, Cw5, Cw7, DR3, DR4, DR9 and DQ3 were decreased. HLA A25 (10), B54 (22), B56 (22), B39 (16), B57 (17), split antigens along with B14 and Cw8 were not identified in the Marathas.

Table 2 present the two locus haplotype frequencies and linkage disequilibrium of Marathas. It is interesting to note that haplotypes A10-B8, A1-B17, A24-B52, B5-Cw9, B13-Cw3, B15-Cw2, B35-Cw4, DR2-DQ1, DR5-DQ3 and DR1-DQ9 haplotypes had high delta values and significant T values with positive linkage disequilibrium among the Marathas except A2-B12 which

Table 1: HLA antigen frequencies in Marathas from Mumbai Western India

HLA	%PF	Maxlike Gene Frequency	SEGF
A1	28.37	0.1434	0.0158
A2	30.80	0.1569	0.0165
A3	9.00	0.0430	0.0086
A9	25.26	0.1264	0.0148
A10	8.30	0.0396	0.0083
A11	24.57	0.1227	0.0146
A19	21.11	0.0583	0.0100
A23	0.35	0.0016	0.0017
A24	12.11	0.0583	0.0100
A26	2.77	0.0130	0.0047
A28	12.80	0.0618	0.0103
A29	3.11	0.0146	0.0050
A30	0.35	0.0016	0.0017
A31	1.73	0.0081	0.0037
A32	0.35	0.0016	0.0017
A33	2.42	0.0114	0.0044
A36	0.35	0.0016	0.0017
A-	16.26	0.0899	0.0125
B5	17.30	0.0868	0.0123
B7	23.88	0.1221	0.0145
B8	6.23	0.0303	0.0072
B12	10.03	0.0493	0.0092
B13	5.88	0.0286	0.0070
B15	5.19	0.0252	0.0068
B17	12.80	0.0634	0.0105
B18	2.42	0.0117	0.0045
B21	4.50	0.0218	0.0061
B22	3.81	0.0184	0.0056
B27	11.76	0.0581	0.0100
B35	26.30	0.1355	0.0153
B37	5.19	0.0252	0.0066
B38	0.35	0.0016	0.0017
B40	20.42	0.1033	0.0134
B44	8.65	0.0424	0.0086
B51	4.84	0.0235	0.0064
B52	3.46	0.0167	0.0054
B53	3.81	0.0184	0.0056
B55	1.38	0.0067	0.0034
B58	0.69	0.0033	0.0024
B60	2.77	0.0133	0.0048
B61	6.92	0.0337	0.0076
B62	0.69	0.0033	0.0024
B63	0.35	0.0017	0.0017
B73	0.35	0.0017	0.0017
B-	9.69	0.0524	0.0095
Cw1	7.60	0.0390	0.0150
Cw2	4.30	0.0220	0.0110
Cw3	29.30	0.1590	0.0290
Cw4	19.60	0.1030	0.0240
Cw5	1.10	0.0050	0.0050
Cw6	20.70	0.1090	0.0240
Cw7	3.30	0.0160	0.0090
Cw-	83.70	0.5960	0.0570
DR1	16.84	0.0795	0.0205
DR2	50.53	0.2678	0.0375
DR3	7.37	0.0339	0.0134
DR4	8.42	0.0389	0.0143
DR5	15.79	0.0743	0.0198
DR6	12.63	0.0589	0.0176
DR7	29.47	0.1446	0.0276
DR9	7.37	0.0339	0.0134
DR10	14.74	0.0692	0.0191
DR11	6.32	0.0290	0.0123
DR-	28.42	0.1699	0.0299
DRw52	52.38	0.2452	0.0764
DRw53	42.86	0.1931	0.0678
DRW-	90.48	0.5617	0.1156
DQ1	74.47	0.4015	0.0462
DQ2	28.72	0.1264	0.0259
DQ3	19.15	0.0818	0.0209
DQ-	65.96	0.3903	0.0456

%PF = Percentage Phenotype Frequency
SEGF = standard error of gene frequency

Table 2: Significant haplotype frequencies and significant Linkage disequilibrium identified in Marathas

Haplotype	HF	Delta(Δ)	t value ^a	Ki ²
A1 - B17	28.0	17.2	2.60	8.06
A10-B8	22.4	20.6	3.61	66.42
A24-B52	10.0	9.0	2.27	17.90
A28-B15	11.0	8.9	2.11	9.34
B5-Cw9	21.0	12.9	2.54	7.79
B13-Cw3	15.5	12.3	2.83	17.09
B15-Cw2	26.9	13.9	2.29	5.73
B35-Cw4	10.7	8.3	2.33	9.89
DR1-DQ9	17.1	11.3	2.76	10.27
DR2-DQ1	52.4	21.4	2.88	8.13
DR5-DQ3	18.9	14.4	3.38	22.09
DR7-DQ2	36.4	21.3	3.59	15.58
DR10-DQ1	28.3	16.6	2.14	4.16
A2-B12 ^s	11.0	-15.4	2.34	5.43

HF = Haplotype frequency per 1000.

Delta = Linkage disequilibrium per 1000.

^s = significant Negative Linkage disequilibrium.

^a = t value > 2 indicates positive for Delta.

Ki² = Chisquare value

was the only haplotype identified in negative linkage disequilibrium.

DISCUSSION

Theoretically high polymorphism of a gene can occur due to mutation rate, selection, genetic hitchhiking or a combination of all the three (Kaufmann 1996). In addition data have been used to generate hypothesis about the nature of selective forces operating on the HLA loci to elucidate the pattern of human evolution and migration. Worldwide most populations contain only 20 to 30 alleles in a HLA loci although over 100 alleles have been identified. Earlier population studies have indicated that there are many alleles and haplotypes that appear to be specific for a given population group. Indigenous populations or caste groups show a very restricted diversity of alleles at a particular HLA loci consistent within a population (Apple and Elrich 1996). Moreover specific alleles found uniquely in a particular indigenous group may have been generated by point mutation or gene conversion from the ancestral allele after the group separated from the other groups (Trachtenberg et al. 1995; Rajalingam et al. 1997). Multiple polymorphic alleles in a population are maintained at appreciable frequencies due to overdominance (heterozygous advantage), frequency dependent selection or other selective forces (Titus-

Trachtenberg et al. 1994). Both selective forces and a high rate of germline diversification are involved in the evolution of HLA allelic diversity. A newly arisen favorable variant allele might co-exist with the parental allele rather than replacing it when selective forces favoring diversity is operating. Recently, newer HLA alleles like B78, B5102, and B3506 in South Indians (Tait et al. 1998) and a DR4 novel subtype in North Indians (Mehra et al. 1994) have been identified to co-exist with other alleles.

Population specific distribution of HLA alleles is necessary and interesting both in population genetics and in HLA disease association studies (Bodmer 1987; Bodmer et al. 1999). 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. This kind of study if extensively documented helps in constructing a tree showing the demic relationships between various castes and clans. Together with history, language, social relationships and HLA studies provides a strong evidence of genetic relationships between these castes and clans in addition to social relationships. Anthropological studies have demonstrated that the distribution of HLA alleles differs from one ethnic group to another and more novel alleles may be discovered in future population studies (Munkhbhat et al. 1997). These studies suggest that majority of the North Indian populations possess Caucasian characteristics, but the typical Caucasian haplotype A1-B8 is yet to be observed, on the other hand A10-B8 is commonly found in most of the Indian studies reported (Agrawal et al. 1999; Shankarkumar et al. 1999; Chhaya et al. 2000), Turks (Svejgard et al. 1972), Middle East (Seth et al. 1985) and West Pakistan (Soleheim et al. 1972). It might be possible that founder effect may be the important cause for this pattern of distribution in haplotypes.

Thus, the present study reveals, 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. One of the important aspects of the present study is the utilization of anti HLA antibodies obtained from indigenous population in parallel with commercial antisera. However, the real micro-dissection of the

population will be possible when such studies are carried out at the molecular level.

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