TECHNICAL NOTE

Mohammad A. Tahir,[1] Ph.D.; Joseph Caruso,[1] B.S.; Bruce Budowle,[2] Ph.D.; Nasir Aziz,[3] M.D.; and Gabriel E. Novick,[1] M.D., Ph.D.

Distribution of HLA-DQα and Polymarker (LDLR, GC, GYPA, HBGG, and D7S8) Alleles in Arab and Pakistani Populations Living in Abu Dhabi, United Arab Emirates

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ABSTRACT: Randomly collected blood samples from 100 Arabs and 100 Pakistanis residing in Abu Dhabi were analyzed using the HLA-DQα and polymarker (LDLR, GC, GYPA, HBGG, D7S8) PCR based reverse dot blot systems. Allelic frequencies for each allele and observed heterozygosity for each locus were calculated. Departures from Hardy-Weinberg expectations (HWE) were determined using the unbiased estimate of the expected homozygote/heterozygote frequencies, the likelihood ratio test and the exact test. No significant departures from HWE expectations were detected.

KEYWORDS: forensic science, DNA typing, polymerase chain reaction, HLA-DQA1, LDLR, GC, GYPA, HBGG, D7S8, polymarker, population genetics, allele frequencies, Arab, Pakistani

TABLE 1—*Observed allele frequencies for PM loci of Arab and Pakistani individuals living in Abu Dhabi, United Arab Emirates (UAE).*

Locus	Allele	Arab (n = 100)	Pakistani (n = 100)
LDLR	A	0.435	0.450
	B	0.560	0.550
GYPA	A	0.730	0.610
	B	0.270	0.390
HBGG	A	0.345	0.535
	B	0.605	0.455
	C	0.050	0.010
D7S8	A	0.630	0.740
	B	0.370	0.260
GC	A	0.200	0.240
	B	0.275	0.205
	C	0.525	0.555

n = number of individuals typed.

The discovery of restriction fragment length polymorphism (1), has revolutionized the analysis of biological evidence in forensic sciences. Since then, RFLP analysis has been the backbone of the DNA typing field because it is reliable and reproducible, although it is labor intensive and limited by both quantity and quality of DNA sample.

Polymerase chain reaction (PCR) based strategies, on the contrary, are less limited by quantity or quality of the sample (2) are fast, easy to perform, highly sensitive, and use nonisotopic assays.

HLA-DQα and polymarker (LDLR, GC, GYPA, HBGG, D7S8) are some of the best characterized PCR-based forensic systems (2). Both systems are based on the reverse dot blot format and the use of sequence specific oligonucleotide (SSO) probes. To interpret the significance of PCR results in a forensic context, and as with any other method, the distribution of the different allelic variants for the various loci should be studied

In this study we analyzed the distribution of the HLA-DQα and polymarker (LDLR, GC, GYPA, HBGG, D7S8) alleles in individuals from Arab and Pakistani populations residing in Abu Dhabi, United Arab Emirates.

Methods

Blood samples were randomly collected in EDTA Vacutainer tubes from 100 Arabs and 100 Pakistanis, all unrelated, from the population of Abu Dhabi. Blood stains were prepared on sterilized cotton cloth.

DNA was extracted according to the FBI protocol, with slight modifications (3,4). PCR amplification of HLA-DQα and polymarker was carried out using the Perkin Elmer AmpliType HLA DQ-α and AmpliType PM amplification and typing kits in a Perkin Elmer 480 Thermal Cycler according to manufacturer's recommendations. Both Arab and Pakistani groups were characterized with polymarker, while only Pakistani samples were typed for HLA-DQα.

Frequencies for each allele were calculated directly from the genotypic classes in the sample set. Expected heterozygosity was calculated and possible departures from Hardy-Weinberg expectations (HWE) were determined using the unbiased estimate of the

[1]Indianapolis-Marion County Forensic Services Agency, Indianapolis, IN.
[2]Forensic Science Research and Training Center, FBI Academy, Quantico, VA.
[3]King Edward Medical College, Lahore, Pakistan.
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TABLE 2—*Observed allele frequencies for HLA-DQα locus in 100 Pakistani individuals from Abu Dhabi, UAE.*

Locus	Allele	Pakistani (n = 100)
DQα	1.1	0.180
	1.2	0.297
	1.3	0.160
	2	0.105
	3	0.110
	4	0.315

n = number of individuals typed.

TABLE 3—*Observed genotype frequencies for PM loci in 100 Arab and 100 Pakistani samples from Abu Dhabi, UAE.*

	Arab (n = 100)						Pakistani (n = 100)					
Loci	AA	AB	BB	AC	BC	CC	AA	AB	BB	AC	BC	CC
LDLR	0.19	0.32	0.49	NA	NA	NA	0.20	0.50	0.30	NA	NA	NA
GYPA	0.55	0.09	0.36	NA	NA	NA	0.36	0.50	0.14	NA	NA	NA
HBGG	0.12	0.41	0.37	0.04	0.06	0.0	0.32	0.43	0.24	0.00	0.00	0.01
D7S8	0.40	0.46	0.14	NA	NA	NA	0.58	0.32	0.10	NA	NA	NA
GC	0.03	0.10	0.10	0.24	0.25	0.28	0.06	0.07	0.05	0.29	0.24	0.29

NA = There is no C allele on the strips provided with the AmpliType PM PCR Amplification and Typing Kit for LDLR, GYPA, and D7S8.

LDLR: Arab: Observed homozygosity = 0.51; expected homozygosity (unbiased) = 0.51; observed heterozygosity = 0.49; expected heterozygosity (unbiased) = 0.49; Homozygosity test (p = 0.94); likelihood ratio test (p = 0.98); exact test (p = 1.00).
Pakistani: Observed homozygosity = 0.50; expected homozygosity (unbiased) = 0.50; observed heterozygosity = 0.50; expected heterozygosity (unbiased) = 0.50; Homozygosity test (p = 0.96); likelihood ratio test (p = 0.92); exact test (p = 1.00).

GYPA: Arab: Observed homozygosity = 0.64; expected homozygosity (unbiased) = 0.60; observed heterozygosity = 0.36; expected heterozygosity (unbiased) = 0.39; Homozygosity test (p = 0.46); likelihood ratio test (p = 0.39); exact test (p = 0.47).
Pakistani: Observed homozygosity = 0.50; expected homozygosity (unbiased) = 0.52; observed heterozygosity = 0.50; expected heterozygosity (unbiased) = 0.48; Homozygosity test (p = 0.66); likelihood ratio test (p = 0.61); exact test (p = 0.68).

HBGG: Arab: Observed homozygosity = 0.49; expected homozygosity (unbiased) = 0.49; observed heterozygosity = 0.51; expected heterozygosity (unbiased) = 0.51; Homozygosity test (p = 0.92); likelihood ratio test (p = 0.90); exact test (p = 0.95).
Pakistani: Observed homozygosity = 0.57; expected homozygosity (unbiased) = 0.50; observed heterozygosity = 0.43; expected heterozygosity (unbiased) = 0.50; Homozygosity test (p = 0.16); likelihood ratio test (p = 0.17); exact test (p = 0.17).

D7S8: Arab: Observed homozygosity = 0.54; expected homozygosity (unbiased) = 0.53; observed heterozygosity = 0.46; expected heterozygosity (unbiased) = 0.47; Homozygosity test (p = 0.86); likelihood ratio test (p = 0.89); exact test (p = 1.00).
Pakistani: observed homozygosity = 0.68; expected homozygosity (unbiased) = 0.61; observed heterozygosity = 0.32; expected heterozygosity (unbiased) = 0.39; Homozygosity test (p = 0.17); likelihood ratio test (p = 0.10); exact test (p = 0.11).

GC: Arab: Observed homozygosity = 0.41; expected homozygosity (unbiased) = 0.39; observed heterozygosity = 0.59; expected heterozygosity (unbiased) = 0.61; Homozygosity test (p = 0.65); likelihood ratio test (p = 0.56); exact test (p = 0.56).
Pakistani: Observed homozygosity = 0.40; expected homozygosity (unbiased) = 0.40; observed heterozygosity = 0.60; expected heterozygosity (unbiased) = 0.60; Homozygosity test (p = 0.92); likelihood ratio test (p = 0.69); exact test (p = 0.70).

n = number of individuals typed.

TABLE 4—*Observed genotypic distribution for HLA-DQα in 100 Pakistani individuals living in Abu Dhabi, UAE.*

Genotype	Frequency (n = 100)
1.1-1.1	0.04
1.1-1.2	0.04
1.1-1.3	0.09
1.1-2	0.01
1.1-3	0.01
1.1-4	0.13
1.2-1.2	0.03
1.2-1.3	0.03
1.2-2	0.05
1.2-3	0.01
1.2-4	0.07
1.3-1.3	0.01
1.3-2	0.06
1.3-3	0.04
1.3-4	0.08
2-2	0.00
2-3	0.04
2-4	0.05
3-3	0.00
3-4	0.12
4-4	0.09

Observed homozygosity = 0.17; expected homozygosity (unbiased) = 0.19; observed heterozygosity = 0.83; expected heterozygosity (unbiased) = 0.80; Homozygosity test (p = 0.56); likelihood ratio test (p = 0.05); exact test (p = 0.12).
n = number of individuals typed.

expected homozygote/heterozygote frequencies (5), the likelihood ratio test (6–8) and the exact test (9,10).

A 2 × C contingency table exact test was used to generate a G-statistic (1000 shuffling experiments) (11,12) to test for homogeneity between the sample populations and other sample groups.

TABLE 5—*G statistic test (p values) for homogeneity on PM and HLA-DQA1 between Abu Dhabi Arabs, Abu Dhabi Pakistanis and other population groups.*

	Arabs (n = 100)	Pakistani (n = 100)
Arabs LDLR	—	0.830
Arabs GYPA	—	0.012*
Arabs HBGG	—	0.001*
Arabs D7S8	—	0.024*
Arabs GC	—	0.214
Arabs (ref. 14) LDLR	0.704	0.992
Arabs (ref. 14) GYPA	0.022*	0.916
Arabs (ref. 14) HBGG	0.458	0.023*
Arabs (ref. 14) D7S8	0.746	0.064
Arabs (ref. 14) GC	0.913	0.313
Dubai Arabs (ref. 15) LDLR	0.913	0.859
Dubai Arabs (ref. 15) GYPA	0.219	0.121
Dubai Arabs (ref. 15) HBGG	0.007*	10^{-3}*
Dubai Arabs (ref. 15) D7S8	0.848	NA
Dubai Arabs (ref. 15) GC	0.942	0.012*
African American (ref. 13) LDLR	10^{-3}*	10^{-3}*
African American (ref. 13) GYPA	10^{-3}*	0.007
African American (ref. 13) HBGG	10^{-3}*	10^{-3}*
African American (ref. 13) D7S8	0.798	0.004
African American (ref. 13) GC	10^{-3}*	10^{-3}*
US Caucasian (ref. 13) LDLR	0.695	1.000
US Caucasian (ref. 13) GYPA	0.002*	0.586
US Caucasian (ref. 13) HBGG	0.001*	0.344
US Caucasian (ref. 13) D7S8	0.791	0.003
US Caucasian (ref. 13) GC	0.021*	0.669
Arabs (ref. 14) DQA1	0.128	NA
Dubai Arabs (ref. 15) DQA1	10^{-3}*	NA
African American (ref. 13) DQA1	10^{-3}*	NA
US Caucasian (ref. 13) DQA1	10^{-3}*	NA

* = Significant.
NA = Not available.

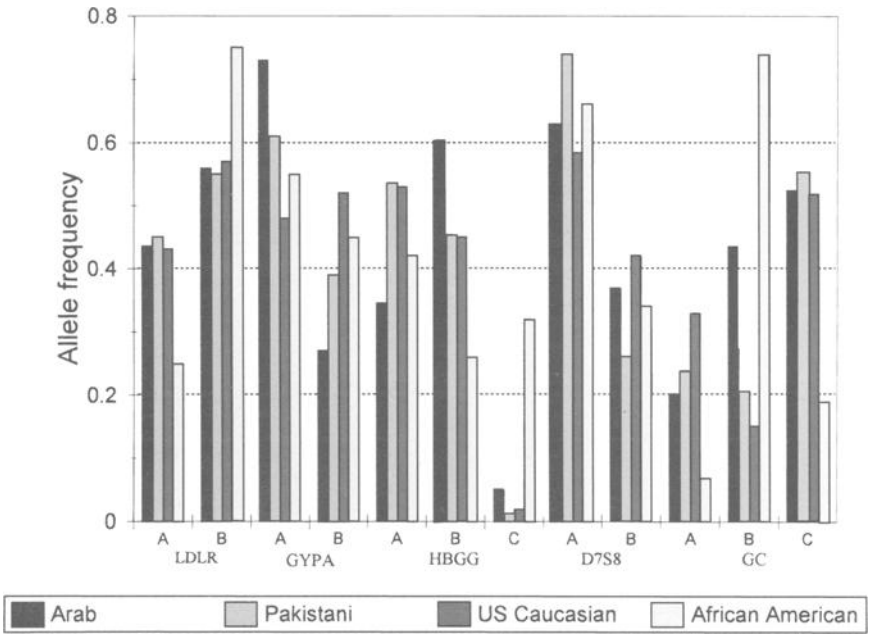

FIG. 1—*Allele frequency distribution for PM loci in Arab and Pakistani individuals from Abu Dhabi, UAE. Allele frequency data on Caucasians and African Americans from the FBI database is also depicted (13).*

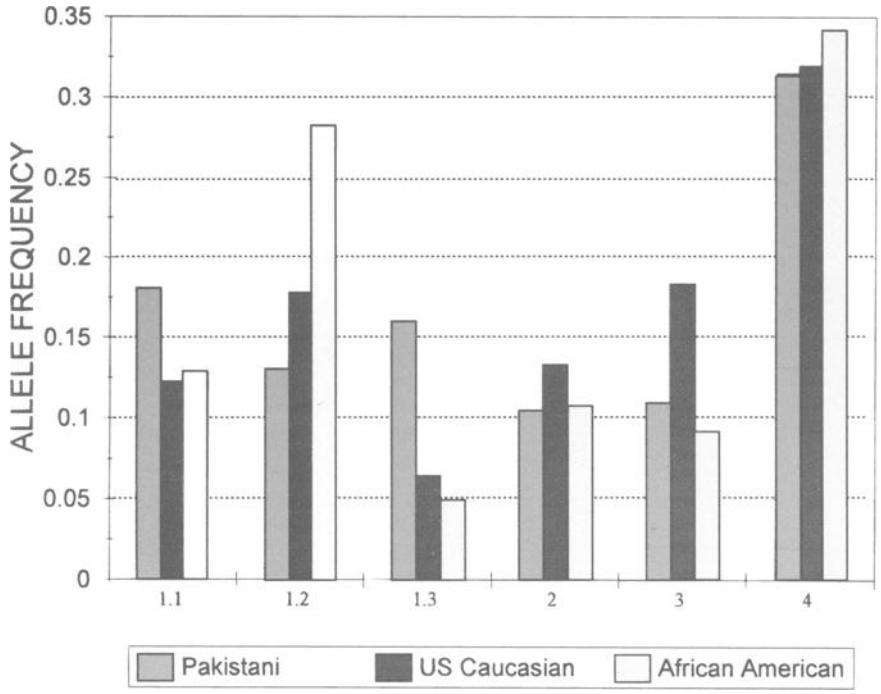

FIG. 2—*Allele frequency distribution for HLA-DQα locus in Pakistani individuals from Abu Dhabi, UAE. Data on allele frequency in Caucasians and African Americans from the FBI database is also represented (13).*

Results and Discussion

Allelic frequencies obtained in the two population sets for the five PM loci and those for HLA-DQα in the Pakistani set are shown in Tables 1 and 2, respectively. The distribution of observed genotypic frequencies for both markers in the groups studied is shown in Tables 3 and 4, respectively. Figures 1 and 2 show the allele frequency distributions for PM and HLA-DQα loci in both populations in relation to the FBI database for Caucasians and African Americans (13).

The observed heterozygosity for all five PM loci, in the Arabs ranged from 36% for GYPA to 59% for Gc. In the Pakistani group, the observed heterozygosity for all five PM loci ranged from 32% for D7S8, to 60% for Gc. Observed heterozygosity for the DQα locus in the Pakistani group was 83%. There were no detectable departures from HWE.

The allele frequencies for Abu Dhabi Arabs are statistically different at three of the five PM loci compared with the Pakistani group (Table 5). Moreover, the Abu Dhabi Arabs tend to have different allele frequency distributions at a number of loci compared with African Americans and US Caucasians. These observations are not surprising for these particular population groups. In contrasts, the majority of loci are similar between Arab groups. This supports the notion that Arab groups are genetically more similar to one another than compared with other population groups. The Pakistanis, while showing similar trends, also show similarities in their allele distributions with US Caucasians.

The above homogeneity analyses are an example of how statistics can detect significant differences between populations, and yet if either population was used as a reference database, the resulting DNA profile frequency would not be substantially different for most practical forensic applications.

The data here support that these databases can be used for estimating the rarity of DNA profile frequencies. Moreover, the data may be useful for population genetics studies.

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Additional information and reprint requests:
Mohammad A. Tahir, Ph.D.
Indianapolis Marion County
Forensic Services Agency
40 S. Alabama St
Indianapolis, IN 46204