Original Article
Diagnostic Immunology

Ann Lab Med 2015;35:429-435
http://dx.doi.org/10.3343/alm.2015.35.4.429
ISSN 2234-3806 • eISSN 2234-3814

ANNALS OF LABORATORY MEDICINE

# Allele and Haplotype Frequencies of Human Leukocyte Antigen-A, -B, -C, -DRB1, and -DQB1 From Sequence-Based DNA Typing Data in Koreans

Ji Won In, M.D.[1], Eun Youn Roh, M.D.[1], Sohee Oh, Ph.D.[2], Sue Shin, M.D.[1], Kyoung Un Park, M.D.[1], and Eun Young Song, M.D.[1,3]

Department of Laboratory Medicine[1], Seoul National University College of Medicine; Department of Biostatistics[2], Seoul National University Boramae Medical Center; Department of Molecular Medicine and Biopharmaceutical Sciences[3], Graduate School of Convergence Science and Technology and College of Medicine, Medical Research Center, Seoul National University, Seoul, Korea

**Background:** Data on allele frequencies (AFs) and haplotype frequencies (HFs) of HLA-C and -DQB1 are limited in Koreans. We investigated AFs and HFs of HLA-A, -B, -C, -DRB1, and -DQB1 in Koreans by high-resolution sequence-based typing (SBT).

**Methods:** Hematopoietic stem cells were obtained from 613 healthy, unrelated donors to analyze HLA-A, -B, -C, -DRB1, and -DQB1 genotypes by using AlleleSEQR HLA-A, -B, -C, -DRB1, and -DQB1 SBT kits (Abbott Molecular, USA), respectively. Alleles belonging to *HLA-C*07:01/07:06* group were further discriminated by using PCR-sequence specific primer analysis. AFs and HFs were calculated by direct counting and maximum likelihood method, respectively.

**Results:** In all, 24 HLA-A, 46 HLA-B, 24 HLA-C, 29 HLA-DRB1, and 15 HLA-DQB1 alleles were identified. AFs and HFs of HLA-A, -B, and -DRB1 were similar to those reported previously. For the HLA-C locus, *C*01:02* was the most common allele, followed by *C*03:03*, *C*03:04*, *C*14:02*, *C*03:02*, and *C*07:02* (AF ≥7%). AFs of *C*07:01* and *C*07:06* were 0.16% and 3.18%, respectively. For the HLA-DQB1 locus, *DQB1*03:01* was the most common allele, followed by *DQB1*03:03*, **03:02*, **06:01*, **05:01*, **04:01*, and **06:02* (AF ≥7%). AFs of *DQB1*02:01* and *DQB1*02:02* were 2.12% and 6.69%, respectively. HFs of *A*33:03-C*07:06* and *C*07:06-B*44:03* were 3.09% and 3.10%, respectively, while those of *DRB1*07:01-DQB1*02:02* and *DRB1*03:01-DQB1*02:01* were 6.61% and 2.04%, respectively.

**Conclusions:** This study reported AFs and HFs of HLA, including HLA-C and -DQB1, in Koreans by using high-resolution SBT. These data can be used to resolve ambiguous results of HLA typing for organ and hematopoietic stem cell transplantations.

**Key Words:** Alleles, Frequency, Haplotypes, HLA antigens, Korea, Sequence analysis

**Received:** December 3, 2014
**Revision received:** December 19, 2014
**Accepted:** April 1, 2015

**Corresponding author:** Eun Young Song
Department of Laboratory Medicine, Seoul National University College of Medicine, 101 Daehak-ro, Jongno-gu, Seoul 110-744, Korea
Tel: +82-2-2072-0197
Fax: +82-2-744-8872
E-mail: eysong1@snu.ac.kr

**© The Korean Society for Laboratory Medicine**
This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/3.0) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

## INTRODUCTION

Number of known alleles has rapidly increased because of the use of DNA-based HLA typing methods. Allele (AF) and haplotype frequencies (HF) of HLA are different among different ethnic groups and regions. In Koreans, AF and HF of HLA are reported by using generic DNA typing [1-4] and high-resolution genotyping [5-7]. However, genotyping data for both of HLA-C and -DQB1 alleles using sequence-based typing (SBT) have not been obtained in previous studies, and some alleles such as *C*07:01/*07:06* and *DQB1*02:01/02:02* have not been discriminated [2-5]. Recent studies have shown that HLA-C and

-DQB1 matches are important for hematopoietic stem cell and organ transplantations [8-12]. Understanding the exact frequencies of HLA and other common, well-defined alleles in different ethnic groups is important to resolve ambiguous results of HLA typing. Higher AF of *C*07:06* in the Chinese than in Caucasians was the cause of PCR dropout of widely used AlleleSEQR HLA-C Plus kit (Abbott Molecular, Des Plaines, IL, USA) because the kit was mainly developed for HLA typing for Caucasians [13]. The present study analyzed the AF and HF of HLA-A, -B, -C, -DRB1, and -DQB1 in Koreans by using high-resolution SBT and compared them with other ethnic groups.

## METHODS

### 1. Subjects

Hematopoietic stem cells were obtained from 613 healthy Korean donors (559 healthy unrelated adults and 54 umbilical cord blood units) for high-resolution HLA typing at Seoul National University Hospital from January 2006 to July 2014. This study was approved by the Institutional Review Board (IRB) of Seoul National University Hospital (IRB No. 1408-028-601).

### 2. SBT analysis

Genomic DNA was extracted from white blood cells obtained from the peripheral blood of the donors and umbilical cord blood units by using QuickGene-Mini80 DNA isolation system (Fujifilm, Tokyo, Japan).

SBT of HLA-A, -B, -C, -DRB1, and -DQB1 was performed by using AlleleSEQR HLA-A, -B, -C, -DRB1, and -DQB1 SBT kits (Abbott Molecular), respectively. For HLA class I alleles (HLA-A, -B, and -C), PCR mixture was prepared using 16 μL of master mix, 80-160 ng of genomic DNA, and 0.3 μL of AmpliTaq Gold polymerase (Abbott Molecular) at a final volume of 10 μL. For HLA class II alleles (HLA-DRB1 and -DQB1), PCR mixture was prepared by using 8 μL of master mix, 40-80 ng of genomic DNA, and 0.1 μL of AmpliTaq Gold polymerase at a final volume of 10 μL. Amplification conditions were as follows: initial denaturation at 95°C for 10 min, followed by 36 cycles of denaturation at 96°C for 20 sec, annealing at 60°C for 30 sec, and elongation at 72°C for 3 min.

Next, 16 μL of the PCR products were mixed with 3 μL of ExoSAP-IT and were purified at 37°C for 15-30 min and 80°C for 15 min. Exons 2, 3, and 4 were sequenced for HLA class I alleles (HLA-A, -B, and -C); exon 2 and codon 86 were sequenced for HLA-DRB1; and exons 2 and 3 were sequenced for HLA-DQB1. Next, 8 μL of sequencing mix was added to 2 μL of the purified PCR products, and sequencing was performed by using 25 thermal cycles of denaturation at 96°C for 20 sec, annealing at 50°C for 30 sec, and elongation at 60°C for 2 min. Next, 2 μL of sodium acetate/EDTA buffer and 25 μL of absolute ethanol (EtOH) were added to the mixture containing sequenced products. The mixture was then vigorously vortexed and centrifuged at 2,000*g* for 30 min, and the supernatant was removed. Next, 50 μL of 80% EtOH was added, and the mixture was centrifuged at 2,000*g* for 5 min; this process was repeated twice. Finally, 15 μL of highly deionized formamide (Applied Biosystems, Foster City, CA, USA) and 15 μL of 0.3 mM EDTA were added, and the mixture was loaded onto 3730xl DNA Analyzer (Applied Biosystems, Foster City, CA, USA).

The obtained electropherograms were processed by using Assign SBT (Conexio Genomics, Fremantle Western Australia, Australia). To resolve ambiguity in *C*07:01/07:06/07:18* group, exon 5 sequences of HLA-C were further analyzed, as described previously [14].

### 3. Statistical analysis

AFs and HFs of the HLA alleles were determined by using direct counting and maximum likelihood method, respectively. For 2-locus haplotypes, relative linkage disequilibrium (RLD) values between two alleles at different loci were analyzed and chi-square values were calculated for statistical significance. HFs of HLA A-C-B-DRB1-DQB1 haplotypes and AFs of HLA-C and -DQB1 in Koreans were compared with those in other populations by using chi-square test or Fisher's exact test, as appropriate. Bonferroni correction was applied by multiplying *P* values with the number of comparisons made (24 for HLA A-C-B-DRB1-DQB1 haplotypes, 24 for HLA-C, and 14 for HLA-DQB1) to calculate corrected *P* values (Pc). *Pc* value of $<0.05$ was considered significantly different. All analyses were performed by using R version 3.1.2 software with gap package (http://cran.us.r-project.org/) [15].

## RESULTS

### 1. AFs of HLA -A, -B, -C, -DRB1, and -DQB1

AFs of HLA classes I and II are listed in Table 1. For the HLA-A locus, A2 group comprised more than 50% of all the HLA-A alleles. In all, 24 distinct alleles were identified for the HLA-A locus. Of these, *A*24:02* was the most common allele, followed by *A*02:01*, *A*33:03*, *A*02:06*, and *A*11:01* (AF ≥5%). The HLA-B locus showed the greatest diversity, with 46 alleles. Of these, *B*51:01*, *B*15:01*, and *B*44:03* were the most com-


http://dx.doi.org/10.3343/alm.2015.35.4.429

**Table 1.** AFs (%) of HLA-A, -B, -C, -DRB1, and -DQB1 in Koreans (N=613)

| Allele | AF (%) | Allele | AF (%) | Allele | AF (%) |
|---|---|---|---|---|---|
| *A*01:01* | 1.88 | *B*07:02* | 3.10 | ***DRB1*01:01*** | **6.44** |
| ***A*02:01*** | **17.62** | *B*07:05*† | 0.41 | *DRB1*03:01* | 2.12 |
| *A*02:03* | 0.73 | *B*08:01* | 0.33 | *DRB1*04:01* | 0.98 |
| ***A*02:06*** | **9.46** | *B*13:01* | 2.28 | *DRB1*04:03* | 3.10 |
| *A*02:07* | 3.51 | *B*13:02* | 4.16 | *DRB1*04:04* | 1.96 |
| *A*02:10* | 0.33 | *B*14:01* | 1.96 | ***DRB1*04:05*** | **9.05** |
| *A*03:01* | 1.55 | ***B*15:01*** | **8.73** | *DRB1*04:06* | 4.16 |
| *A*03:02* | 0.08 | *B*15:02* | 0.24 | *DRB1*04:07* | 0.49 |
| ***A*11:01*** | **7.91** | *B*15:07* | 1.47 | *DRB1*04:10* | 1.39 |
| *A*11:02* | 0.49 | *B*15:11* | 1.55 | ***DRB1*07:01*** | **6.93** |
| *A*23:01* | 0.08 | *B*15:17* | 0.08 | *DRB1*08:02* | 1.96 |
| ***A*24:02*** | **22.68** | *B*15:18* | 1.06 | ***DRB1*08:03*** | **6.61** |
| *A*24:08* | 0.08 | *B*15:27* | 0.33 | ***DRB1*09:01*** | **9.46** |
| *A*24:20* | 0.24 | *B*15:38* | 0.16 | *DRB1*10:01* | 1.96 |
| *A*26:01* | 2.61 | *B*27:04* | 0.16 | *DRB1*11:01* | 3.83 |
| *A*26:02* | 2.28 | *B*27:05* | 3.10 | *DRB1*11:06* | 0.08 |
| *A*26:03* | 0.82 | ***B*35:01*** | **6.12** | *DRB1*12:01*‡ | 4.40 |
| *A*29:01* | 0.33 | *B*35:03* | 0.16 | *DRB1*12:02* | 3.18 |
| *A*30:01* | 3.92 | *B*35:31* | 0.08 | *DRB1*13:01* | 1.63 |
| *A*30:04* | 2.12 | *B*37:01* | 2.12 | ***DRB1*13:02*** | **9.30** |
| *A*31:01* | 4.57 | *B*38:01* | 0.08 | *DRB1*13:07* | 0.08 |
| *A*32:01* | 0.57 | *B*38:02* | 0.82 | *DRB1*14:03* | 1.22 |
| ***A*33:03*** | **16.07** | *B*39:01* | 0.90 | *DRB1*14:05* | 3.59 |
| *A*33:25* | 0.08 | *B*40:01* | 3.83 | *DRB1*14:06* | 0.73 |
| | | *B*40:02* | 4.24 | *DRB1*14:07* | 0.08 |
| ***C*01:02*** | **17.81** | *B*40:03* | 0.57 | *DRB1*14:54* | 2.45 |
| *C*01:03* | 0.08 | *B*40:06* | 3.59 | ***DRB1*15:01*** | **8.24** |
| *C*01:04* | 0.08 | *B*40:73* | 0.08 | *DRB1*15:02* | 3.75 |
| *C*02:02* | 0.49 | *B*44:02* | 1.47 | *DRB1*16:02* | 0.82 |
| ***C*03:02*** | **7.42** | ***B*44:03*** | **8.16** | | |
| ***C*03:03*** | **11.83** | ***B*46:01*** | **5.06** | *DQB1*02:01* | 2.12 |
| ***C*03:04*** | **8.97** | *B*47:01* | 0.08 | ***DQB1*02:02*** | **6.69** |
| *C*04:01* | 4.89 | *B*48:01* | 3.91 | ***DQB1*03:01*** | **14.03** |
| *C*05:01* | 1.47 | *B*50:01* | 0.08 | ***DQB1*03:02*** | **9.62** |
| ***C*06:02*** | **6.36** | ***B*51:01*** | **9.54** | ***DQB1*03:03*** | **11.17** |
| *C*07:01* | 0.16 | *B*51:02* | 0.33 | ***DQB1*04:01*** | **8.81** |
| ***C*07:02*** | **7.26** | *B*52:01* | 2.85 | *DQB1*04:02* | 3.92 |
| *C*07:04* | 0.90 | ***B*54:01*** | **5.71** | ***DQB1*05:01*** | **8.97** |
| *C*07:06* | 3.18 | *B*55:02* | 1.31 | *DQB1*05:02* | 2.12 |
| ***C*08:01*** | **6.93** | *B*55:04* | 0.16 | *DQB1*05:03* | 4.49 |
| *C*08:02* | 1.96 | *B*55:07* | 0.08 | ***DQB1*06:01*** | **9.38** |
| *C*08:03* | 0.98 | *B*56:01* | 0.08 | ***DQB1*06:02*** | **7.75** |
| *C*12:02* | 3.10 | *B*57:01* | 0.08 | *DQB1*06:03* | 1.63 |
| *C*12:03* | 0.33 | ***B*58:01*** | **6.77** | ***DQB1*06:04*** | **5.06** |
| ***C*14:02*** | **7.99** | *B*59:01* | 1.71 | *DQB1*06:09* | 4.24 |
| ***C*14:03*** | **5.14** | *B*67:01* | 0.90 | | |
| *C*15:02* | 2.20 | | | | |
| *C*15:05* | 0.41 | | | | |
| *C*16:02* | 0.08 | | | | |

AFs of ≥5% are boldfaced.
†*B*07:05/07:06* and ‡*DRB1*12:01/12:06/12:10* were not discriminated in the present study.
Abbreviation: AF, allele frequency.

mon alleles, followed by *B*58:01*, *B*35:01*, *B*54:01*, and *B*46:01* (AF ≥5%). In all, 24 alleles were identified for the HLA-C locus. Of these, *C*01:02* was the most common allele, followed by *C*03:03*, *C*03:04*, *C*14:02*, *C*03:02*, *C*07:02*, *C*08:01*, *C*06:02*, and *C*14:03* (AF ≥5%). AFs of *C*07:01* and *C*07:06* were 0.16% and 3.18%, respectively. In all, 29 alleles were identified for the HLA-DRB1 locus. Of these, *DRB1*09:01* was the most common allele, followed by *DRB1*13:02*, *DRB1*04:05*, *DRB1*15:01*, *DRB1*07:01*, *DRB1*08:03*, and *DRB1*01:01* (AF ≥5%). In all, 15 alleles were identified for the HLA-DQB1 locus. The sum of AFs of *DQB1*03:01* and *DQB1*03:03* were 25.2%. AFs of three DQ6 alleles (*DQB1*06:01*, *DQB1*06:02*, and *DQB1*06:04*), *DQB1*02:02*, *DQB1*03:02*, *DQB1*04:01*, and *DQB1*05:01* were ≥5%. AFs of *DQB1*02:01* and *DQB1*02:02* were 2.12% and 6.69%, respectively.

### 2. HFs of HLA -A, -B, -C, -DRB1, and -DQB1

The 2-locus haplotypes exhibiting HFs higher than 0.9% are listed in Table 2. HFs of 31 A-C, 27 B-DRB1, 29 C-B, and 24 DRB1-DQB1 haplotypes were >0.9%. In each set, *A*33:03-C*03:02*, *B*44:03-DRB1*13:02*, *C*14:02-B*51:01*, and *DRB1*09:01-DQB1*03:03* were the most common alleles. Three haplotypes (*C*08:02-B*14:01*, *C*05:01-B*44:02*, and *DRB1*13:01-DQB1*06:03*) showed the strongest associations with RLD values of >0.99. HFs (RLD, $\chi^2$) of *A*33:03-C*07:06* and *C*07:06-B*44:03* were 3.09% (0.40, 83.6) and 3.10% (0.59, 301.3), respectively, while those of *DRB1*07:01-DQB1*02:02* and *DRB1*03:01-DQB1*02:01* were 6.61% (0.97, 377.8) and 2.04% (0.96, 320.0), respectively. Table 3 lists 5-locus HLA A-C-B-DRB1-DQB1 haplotypes having HFs of >0.5%. The most common 5-locus haplotype was *A*33:03-C*14:03-B*44:03-DRB1*13:02-DQB1*06:04*, followed by *A*33:03-C*03:02-B*58:01-DRB1*13:02-DQB1*06:09* (Table 3). HF of *A*33:03-C*07:06-B*44:03-DRB1*07:01-DQB1*02:02* was 2.36%. HF of *A*33:03-C*03:02-B*58:01-DRB1*13:02-DQB1*06:09* was higher while that of *A*24:02-C*12:02-B*52:01-DRB1*15:02-DQB1*06:01* was lower than that in the Japanese (*Pc*<0.01) (Table 3).

### 3. Comparison of AFs of HLA-C and -DQ with other ethnic groups

For the HLA-C locus, AFs of *C*03:02* and *C*07:01/06* were higher and that of *C*12:02* was lower than those in the Japanese (*Pc*<0.001; Table 4). AF of *C*03:03* was higher and that of *C*07:02* was lower than that in the Chinese (*Pc*<0.001). AFs of *C*01:02*, *C*03:02*, *C*08:01*, *C*14:02*, and *C*14:03* were

**Table 2.** HFs (%) of two-locus HLA haplotypes in Koreans†

| Haplotype | | HF (%) | RLD | $\chi^2$ | Haplotype | | HF (%) | RLD | $\chi^2$ |
|---|---|---|---|---|---|---|---|---|---|
| *A*33:03* | *C*03:02* | 6.52 | 0.55 | 162.3 | ***C*14:02*** | ***B*51:01*** | **7.99** | **0.91** | **810.9** |
| *A*02:01* | *C*01:02* | 5.85 | 0.19 | 18.1 | ***C*03:02*** | ***B*58:01*** | **6.77** | **0.95** | **892.4** |
| *A*33:03* | *C*14:03* | 5.05 | 0.52 | 136.9 | *C*01:02* | *B*54:01* | 5.62 | 0.52 | 239.5 |
| *A*24:02* | *C*01:02* | 4.14 | 0.01 | 0.0 | ***C*14:03*** | ***B*44:03*** | **5.05** | **0.77** | **545.3** |
| ***A*30:01*** | ***C*06:02*** | **3.75** | **0.74** | **243.6** | *C*01:02* | *B*46:01* | 4.77 | 0.46 | 210.1 |
| *A*24:02* | *C*03:03* | 3.53 | 0.06 | 1.5 | ***C*06:02*** | ***B*13:02*** | **4.16** | **0.80** | **561.7** |
| *A*24:02* | *C*03:04* | 3.44 | 0.12 | 6.5 | *C*03:03* | *B*35:01* | 3.59 | 0.37 | 132.8 |
| *A*33:03* | *C*07:06* | 3.09 | 0.40 | 83.6 | *C*04:01* | *B*15:01* | 3.34 | 0.48 | 229.4 |
| *A*02:07* | *C*01:02* | 2.82 | 0.31 | 45.5 | *C*07:06* | *B*44:03* | 3.10 | 0.59 | 301.3 |
| *A*02:01* | *C*03:03* | 2.82 | 0.06 | 1.4 | *C*03:03* | *B*15:01* | 3.07 | 0.22 | 55.5 |
| *A*24:02* | *C*14:02* | 2.64 | 0.07 | 3.2 | *C*03:04* | *B*40:02* | 3.03 | 0.46 | 163.0 |
| *A*24:02* | *C*07:02* | 2.63 | 0.09 | 3.3 | *C*07:02* | *B*07:02* | 2.93 | 0.60 | 351.8 |
| *A*24:02* | *C*12:02* | 2.60 | 0.26 | 38.6 | *C*08:01* | *B*40:06* | 2.82 | 0.54 | 308.4 |
| *A*02:06* | *C*01:02* | 2.06 | 0.03 | 0.6 | ***C*12:02*** | ***B*52:01*** | **2.77** | **0.93** | **769.9** |
| *A*02:01* | *C*03:04* | 2.01 | 0.04 | 0.5 | *C*01:02* | *B*27:05* | 2.41 | 0.28 | 60.8 |
| ***A*30:04*** | ***C*08:02*** | **1.96** | **0.96** | **468.9** | *C*08:01* | *B*48:01* | 2.12 | 0.37 | 153.7 |
| *A*02:06* | *C*14:02* | 1.89 | 0.14 | 13.0 | ***C*08:02*** | ***B*14:01*** | **1.96** | **1.00** | **933.2** |
| *A*11:01* | *C*04:01* | 1.75 | 0.23 | 32.7 | *C*06:02* | *B*37:01* | 1.95 | 0.52 | 277.1 |
| *A*11:01* | *C*01:02* | 1.67 | 0.03 | 0.2 | *C*03:04* | *B*40:01* | 1.90 | 0.28 | 72.4 |
| *A*02:06* | *C*08:01* | 1.62 | 0.13 | 10.5 | *C*03:04* | *B*13:01* | 1.85 | 0.38 | 134.6 |
| *A*02:06* | *C*03:03* | 1.46 | 0.04 | 0.6 | *C*01:02* | *B*59:01* | 1.71 | 0.28 | 78.3 |
| *A*31:01* | *C*14:02* | 1.43 | 0.19 | 18.5 | *C*03:03* | *B*15:11* | 1.55 | 0.34 | 115.2 |
| *A*01:01* | *C*06:02* | 1.36 | 0.37 | 69.0 | ***C*05:01*** | ***B*44:02*** | **1.47** | **1.00** | **774.1** |
| *A*02:01* | *C*08:01* | 1.31 | 0.01 | 0.0 | *C*03:03* | *B*15:07* | 1.38 | 0.31 | 109.5 |
| *A*24:02* | *C*08:01* | 1.27 | -0.03 | 0.5 | *C*03:04* | *B*40:06* | 1.08 | 0.14 | 16.9 |
| *A*11:01* | *C*07:02* | 1.22 | 0.09 | 3.9 | *C*08:01* | *B*35:01* | 1.02 | 0.10 | 7.8 |
| *A*02:01* | *C*07:02* | 1.18 | -0.01 | 0.3 | *C*01:02* | *B*55:02* | 1.01 | 0.18 | 28.0 |
| *A*02:01* | *C*14:02* | 1.17 | -0.02 | 0.3 | *C*08:03* | *B*48:01* | 0.98 | 0.49 | 204.0 |
| *A*26:01* | *C*03:03* | 1.14 | 0.16 | 16.5 | *C*07:02* | *B*67:01* | 0.90 | 0.34 | 98.2 |
| *A*02:01* | *C*06:02* | 1.00 | -0.01 | 0.4 | | | | | |
| *A*24:02* | *C*04:01* | 0.91 | -0.02 | 0.3 | ***DRB1*09:01*** | ***DQB1*03:03*** | **9.46** | **0.91** | **360.9** |
| | | | | | ***DRB1*04:05*** | ***DQB1*04:01*** | **8.81** | **0.99** | **384.4** |
| *B*44:03* | *DRB1*13:02* | 4.75 | 0.50 | 303.2 | ***DRB1*15:01*** | ***DQB1*06:02*** | **7.75** | **0.97** | **359.6** |
| *B*58:01* | *DRB1*13:02* | 3.83 | 0.44 | 234.7 | ***DRB1*07:01*** | ***DQB1*02:02*** | **6.61** | **0.97** | **377.8** |
| *B*13:02* | *DRB1*07:01* | 3.65 | 0.66 | 485.5 | ***DRB1*01:01*** | ***DQB1*05:01*** | **6.36** | **0.83** | **263.0** |
| *B*46:01* | *DRB1*08:03* | 3.54 | 0.59 | 461.4 | ***DRB1*08:03*** | ***DQB1*06:01*** | **6.19** | **0.77** | **227.6** |
| *B*15:01* | *DRB1*04:06* | 2.94 | 0.46 | 325.9 | ***DRB1*13:02*** | ***DQB1*06:04*** | **5.06** | **0.72** | **223.0** |
| *B*54:01* | *DRB1*04:05* | 2.92 | 0.36 | 132.5 | *DRB1*13:02* | *DQB1*06:09* | 4.24 | 0.66 | 170.9 |
| *B*07:02* | *DRB1*01:01* | 2.85 | 0.63 | 509.6 | *DRB1*04:06* | *DQB1*03:02* | 4.07 | 0.62 | 172.4 |
| *B*44:03* | *DRB1*07:01* | 2.79 | 0.32 | 123.3 | *DRB1*11:01* | *DQB1*03:01* | 3.65 | 0.47 | 80.7 |
| ***B*52:01*** | ***DRB1*15:02*** | **2.77** | **0.84** | **768.7** | *DRB1*12:01*‡ | *DQB1*03:01* | 3.49 | 0.40 | 59.7 |
| *B*27:05* | *DRB1*01:01* | 2.59 | 0.57 | 331.1 | *DRB1*15:02* | *DQB1*06:01* | 3.15 | 0.50 | 100.1 |
| *B*40:06* | *DRB1*09:01* | 2.09 | 0.32 | 133.2 | *DRB1*04:03* | *DQB1*03:02* | 3.10 | 0.55 | 125.4 |
| ***B*37:01*** | ***DRB1*10:01*** | **1.96** | **0.96** | **1046.6** | *DRB1*12:02* | *DQB1*03:01* | 3.00 | 0.42 | 63.0 |
| ***B*13:01*** | ***DRB1*12:02*** | **1.95** | **0.72** | **637.9** | ***DRB1*14:05*** | ***DQB1*05:03*** | **2.93** | **0.73** | **201.0** |
| *B*58:01* | *DRB1*03:01* | 1.78 | 0.45 | 227.2 | ***DRB1*03:01*** | ***DQB1*02:01*** | **2.04** | **0.96** | **320.0** |
| *B*51:01* | *DRB1*09:01* | 1.57 | 0.08 | 7.3 | *DRB1*10:01* | *DQB1*05:01* | 1.96 | 0.45 | 77.2 |
| ***B*14:01*** | ***DRB1*04:04*** | **1.55** | **0.79** | **627.8** | *DRB1*04:04* | *DQB1*04:02* | 1.71 | 0.61 | 120.4 |
| *B*59:01* | *DRB1*04:05* | 1.54 | 0.37 | 132.2 | ***DRB1*13:01*** | ***DQB1*06:03*** | **1.63** | **1.00** | **409.1** |
| *B*35:01* | *DRB1*15:01* | 1.52 | 0.15 | 31.0 | *DRB1*14:54* | *DQB1*05:03* | 1.35 | 0.39 | 46.8 |
| *B*15:01* | *DRB1*15:01* | 1.30 | 0.08 | 6.5 | *DRB1*04:10* | *DQB1*04:02* | 1.30 | 0.55 | 88.0 |
| *B*54:01* | *DRB1*15:01* | 1.30 | 0.13 | 16.4 | *DRB1*14:03* | *DQB1*03:01* | 1.22 | 0.28 | 25.9 |
| *B*15:07* | *DRB1*04:03* | 1.22 | 0.56 | 371.1 | *DRB1*14:54* | *DQB1*05:02* | 1.13 | 0.48 | 72.5 |
| *B*15:01* | *DRB1*04:05* | 1.22 | 0.05 | 1.9 | *DRB1*08:02* | *DQB1*03:02* | 0.98 | 0.19 | 14.3 |
| *B*51:01* | *DRB1*15:01* | 1.19 | 0.05 | 2.2 | | | | | |
| *B*51:01* | *DRB1*12:01*‡ | 1.07 | 0.11 | 17.2 | | | | | |
| *B*40:01* | *DRB1*04:05* | 1.05 | 0.13 | 13.2 | | | | | |
| *B*40:02* | *DRB1*09:01* | 1.05 | 0.11 | 12.5 | | | | | |
| *B*40:01* | *DRB1*09:01* | 0.97 | 0.11 | 11.8 | | | | | |

Haplotypes with RLD values of ≥0.7 are boldfaced.
†Two-locus HLA haplotypes with HFs of >0.9% are listed; ‡*DRB1*12:01/12:06/12:10* was not discriminated in the present study.
Abbreviations: HF, haplotype frequency; RLD, relative linkage disequilibrium.


http://dx.doi.org/10.3343/alm.2015.35.4.429

**Table 3.** HFs (%) of five-locus HLA haplotypes in Koreans[†] and Japanese

| Haplotypes | | | | | HF (%) | | |
|---|---|---|---|---|---|---|---|
| A* | C* | B* | DRB1* | DQB1* | Present study | Koreans [4] | Japanese [16] |
| *33:03* | *14:03* | *44:03* | *13:02* | *06:04* | 4.49 | 4.23 | 4.07 |
| *33:03* | *03:02* | *58:01* | *13:02* | *06:09* | 3.34 | 2.99 | 0.19[‖] |
| *30:01* | *06:02* | *13:02* | *07:01* | *02:02* | 3.26 | 2.68 | 0.39[§] |
| *24:02* | *12:02* | *52:01* | *15:02* | *06:01* | 2.44 | 1.92 | 7.17[‖] |
| *33:03* | *07:06* | *44:03* | *07:01* | *02:02* | 2.36 | 2.99[‡] | 0.00[§] |
| *02:07* | *01:02* | *46:01* | *08:03* | *06:01* | 2.28 | 0.82 | 1.55 |
| *24:02* | *07:02* | *07:02* | *01:01* | *05:01* | 2.16 | 2.89 | 2.71 |
| *02:01* | *01:02* | *27:05* | *01:01* | *05:01* | 1.76 | 1.13 | |
| *30:04* | *08:02* | *14:01* | *04:04* | *04:02* | 1.55 | 1.24 | |
| *11:01* | *04:01* | *15:01* | *04:06* | *03:02* | 1.48 | 1.24 | 1.74 |
| *33:03* | *03:02* | *58:01* | *03:01* | *02:01* | 1.47 | 1.85 | |
| *24:02* | *01:02* | *54:01* | *04:05* | *04:01* | 1.43 | 0.62 | 1.94 |
| *01:01* | *06:02* | *37:01* | *10:01* | *05:01* | 1.13 | 1.03 | 0.39 |
| *24:02* | *01:02* | *59:01* | *04:05* | *04:01* | 1.00 | 1.13 | 0.97 |
| *24:02* | *14:02* | *51:01* | *09:01* | *03:03* | 0.98 | | |
| *02:01* | *03:04* | *13:01* | *12:02* | *03:01* | 0.88 | 0.93 | 0.58 |
| *02:01* | *01:02* | *54:01* | *15:01* | *06:02* | 0.76 | | |
| *11:01* | *01:02* | *54:01* | *04:05* | *04:01* | 0.73 | 0.93 | 1.16 |
| *24:02* | *03:03* | *15:07* | *04:03* | *03:02* | 0.73 | | 0.19 |
| *02:06* | *08:01* | *40:06* | *09:01* | *03:03* | 0.65 | | |
| *03:01* | *05:01* | *44:02* | *13:01* | *06:03* | 0.65 | | 0.39 |
| *24:02* | *03:04* | *13:01* | *12:02* | *03:01* | 0.59 | | |
| *24:02* | *03:04* | *40:02* | *09:01* | *03:03* | 0.57 | | 0.78 |
| *33:03* | *03:02* | *58:01* | *15:01* | *06:02* | 0.57 | | |

[†]Five-locus HLA haplotypes with HFs of >0.5% are listed; [‡]*C*07:01/*07:06* was not discriminated in the present study; [§]*Pc*<0.05; [‖]*Pc*<0.01.
Abbreviations: HF, haplotype frequency; *Pc*, corrected *P* value.

higher and those of *C*02:02*, *C*04:01*, *C*07:01/06*, *C*12:03*, and *C*16:01* were lower than those in Caucasians (*Pc*<0.001; Table 4). For the HLA-DQB1 locus, AF of *DQB1*02:01/02* was higher and that of *DQB1*06:01* was lower than that in the Japanese (*Pc*<0.001; Table 5). AF of *DQB1*05:02* was lower than that in the Chinese (*Pc*<0.001). AFs of *DQB1*04:01*, **05:01*, and **06:01* were higher and those of *DQB1*02:01/02* and *DQB1*06:02* were lower than those in Caucasians (*Pc*<0.001; Table 5).

## DISCUSSION

In this study, we investigated the distribution of AFs and HFs of five HLA loci (HLA-A, -B, -C, -DRB1, and -DQB1) in 613 healthy Korean hematopoietic stem cell donors by using high-resolution DNA typing. AFs of HLA-A, -B, -DRB1, and -DQB1 were similar to those reported in previous studies [1-7]. For the HLA-C locus, AF of *C*03:02* (7.42%) was lower but that of *C*03:04* (8.97%) was higher than that determined by using PCR-sequence specific oligonucleotides (PCR-SSO) and sequencing in a previous study involving 485 healthy Koreans (10.82% for *C*03:02* and 3.82% for *C*03:04*) [4]. However, AFs reported in our study were similar to those obtained by using PCR-SSO and sequence-specific conformational polymorphism in another study involving 474 healthy Koreans (6.3% for *C*03:02* and 9.9% for *C*03:04*) [5]. This discrepancy should be clarified in further studies by performing high-resolution DNA typing in a larger study population.

Compared with the previous two studies [4, 5], we further discriminated *HLA-C*07:01/*07:06* and determined AFs of *C*07:01* (0.16%) and *C*07:06* (3.18%) and HFs of *A*33:03-C*07:06* (3.09%) and *C*07:06-B*44:03* (3.10%), with signifi-

**Table 4.** AFs (%) of HLA-C in Koreans, Japanese, Chinese, and Caucasians

| Serology | HLA-C† | Koreans‡ (N=613) | Japanese [17] (N=371) | Chinese [18] (N=264) | Caucasians [19] (N=558) |
|---|---|---|---|---|---|
| 1 | *01:02 | 17.8 | 14.8 | 17.4 | 3.7** |
| | *01:03 | 0.1 | 0.4 | | |
| | *01:04 | 0.1 | | | |
| 2 | *02:02 | 0.5 | | | 5.8** |
| 3 (10) | *03:02 | 7.4 | 0.4** | 8.9 | 0.4** |
| 3 (9) | *03:03 | 11.8 | 12.1 | 3.8** | 5.5¶ |
| 3 (10) | *03:04 | 9.0 | 13.7 | 11.2 | 6.2 |
| 4 | *04:01 | 4.9 | 4.6 | 4.5 | 13.0** |
| 5 | *05:01 | 1.5 | 0.4 | | 5.6¶ |
| 6 | *06:02 | 6.4 | 1.6‖ | 2.8 | 9.8 |
| 7 | *07:01 | 0.2 | | 1.7§ | 16.1§,** |
| | *07:02 | 7.3 | 14.6¶ | 21.0** | 11.6 |
| | *07:04 | 0.9 | 0.9 | 0.9 | 2.0 |
| | *07:06 | 3.2 | 0.0** | 0.0‖ | |
| 8 | *08:01 | 6.9 | 7.4 | 13.3 | 0.2** |
| | *08:02 | 2.0 | | 0.2 | 3.1 |
| | *08:03 | 1.0 | 2.0 | | 0.1 |
| Blank | *12:02 | 3.1 | 10.5** | 1.7 | 0.8 |
| | *12:03 | 0.3 | | 1.7 | 7.0** |
| | *14:02 | 8.0 | 4.9 | 3.2 | 1.6** |
| | *14:03 | 5.1 | 8.9 | 0.2‖ | 0.0** |
| | *15:02 | 2.2 | 2.7 | 3.8 | 1.9 |
| | *15:05 | 0.4 | | 1.1 | 0.5 |
| | *16:01 | | | | 2.8** |
| | *16:02 | 0.1 | | | 0.4 |

†HLA-C alleles observed in the present study or having AFs of >1.0% in other populations were listed; ‡Present study; §*C*07:01/*07:06* was not discriminated; ‖*Pc*<0.05; ¶*Pc*<0.01; **Pc*<0.001.
Abbreviations: AF, allele frequency; *Pc*, corrected *P* value.

**Table 5.** AFs (%) of HLA-DQB1 in Korean, Japanese, Chinese, and Caucasian populations

| Serology | DQB1† | Korean‡ (N=613) | Japanese [17] (N=371) | Chinese [18] (N=264) | Caucasian [20] (N=250) |
|---|---|---|---|---|---|
| 2 | *02:01 | 2.1 | 0.3§,** | 12.1§ | 26.6§,** |
| | *02:02 | 6.7 | | | |
| 3 (7) | *03:01 | 14.0 | 11.3 | 24.2¶ | 15.4 |
| 3 (8) | *03:02 | 9.6 | 10.8 | 7.0 | 10.4 |
| 3 (9) | *03:03 | 11.2 | 13.5 | 15.0 | 6.8 |
| 4 | *04:01 | 8.8 | 11.5 | 4.0 | 0.0** |
| | *04:02 | 3.9 | 3.9 | 0.8 | 0.8 |
| 5 | *05:01 | 9.0 | 7.5 | 3.4 | 0.2** |
| | *05:02 | 2.1 | 3.5 | 11.0** | 0.8 |
| | *05:03 | 4.5 | 2.7 | 3.0 | 2.6 |
| 6 | *06:01 | 9.4 | 18.3** | 11.2 | 0.2** |
| | *06:02 | 7.8 | 8.2 | 3.8 | 19.6** |
| | *06:03 | 1.6 | 0.7 | 0.9 | 5.0 |
| | *06:04 | 5.1 | 7.3 | 0.8‖ | 2.0 |
| | *06:09 | 4.2 | 0.4¶ | 1.9 | 0.2‖ |

†HLA-DQB1 alleles observed in the present study or having AFs of >1.0% in other populations were listed; ‡Present study; §*DQB1*02:01/*02:02* was not discriminated; ‖*Pc*<0.05; ¶*Pc*<0.01; **Pc*<0.001.
Abbreviations: AF, allele frequency; *Pc*, corrected *P* value.

cant $\chi^2$ values (83.6 and 301.3, respectively). Strong linkage disequilibrium of *C*07:06* with *A*33:03* or *B*44:03* observed in the present study was in accordance with that reported in a previous study involving 118 healthy Koreans [14]. Therefore, most *A*33:03-C*07:01/06* and *C*07:01/06-B*44:03* haplotypes reported previously [4, 5] would be *A*33:03-C*07:06* and *C*07:06-B*44:03* [4, 5]. Further, we discriminated *DQB1*02:01/*02:02* and determined AFs of *DQB1*02:01* (2.12%) and *DQB1*02:02* (6.69%) and HFs of *DRB1*03:01-DQB1*02:01* (2.04%) and *DRB1*07:01-DQB1*02:02* (6.61%), with significant $\chi^2$ values (320.0 and 377.8, respectively), which were similar to those reported in a previous study involving 414 parents of 207 Korean families [2]. For the 5-locus haplotypes, the results of the present study implied that most *A*33:03-C*07:01/06-B*44:03-DRB1*07:01-DQB1*02:01/02* haplotype reported previously, with HF of 2.99% [4], would be *A*33:03-C*07:06-B*44:03-DRB1*07:01-DQB1*02:02*.

In the present study, all the DQB1 alleles had AFs of >1.5%, with no additional DQB1 allele, compared with those reported in previous studies [2, 4], indicating lower polymorphism of the HLA-DQB1 locus in Koreans. Thus, for organ transplantation, HLA-DQB1 alleles might be analyzed by using an intermediate resolution typing kit to determine compatible donors or to define donor-specific HLA antibody.

AFs of HLA-C alleles in different ethnic groups are listed in Table 4. From the perspective of killer cell immunoglobulin-like receptor (KIR) ligand status, the portion of HLA-C1 group (HLA-C alleles expressing Asn80, such as HLA-Cw1, -Cw3, -Cw7, -Cw8, -Cw12, -Cw13, -Cw14, and -Cw16) [21] was higher in Koreans than in Caucasians (84.2% vs. 61.1%). The portion of C2 group (HLA-C alleles expressing Lys80, such as HLA-Cw2, -Cw4, -Cw5, -Cw6, -Cw15, -Cw17, and -Cw18) was lower in Koreans than in Caucasians (15.8% vs. 36.6%). These differences could affect the frequency of KIR ligand mismatch for hematopoietic stem cell transplantation and possibly the clinical outcome. AFs of HLA-DQB1 alleles in different ethnic groups are listed in Table 5.


http://dx.doi.org/10.3343/alm.2015.35.4.429

AF of *DQB1*06:01* was higher in Koreans (9.4%) than in Caucasians (0.2%), which might cause false reaction while using commercial kits developed in the United States [22].

In summary, we determined the AFs and HFs of HLA-A, -B, -C, -DRB1, and -DQB1 in Koreans by using high-resolution SBT. These data could be used to determine exact HLA types in Koreans for organ and hematopoietic stem cell transplantations.

## Authors' Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

## REFERENCES

1. Roh EY, Kim HS, Kim SM, Lim YM, Han BY, Park MH. HLA-A, -B, -DR allele frequencies and haplotypic associations in Koreans defined by generic-level DNA typing. Korean J Lab Med 2003;23:420-30.
2. Song EY, Park MH, Kang SJ, Park HJ, Kim BC, Tokunaga K, et al. HLA class II allele and haplotype frequencies in Koreans based on 107 families. Tissue Antigens 2002;59:475-86.
3. Hwang SH, Oh HB, Yang JH, Kwon OJ, Shin ES. Distribution of HLA-A, B, C allele and haplotype frequencies in Koreans. Korean J Lab Med 2004;24:396-404.
4. Lee KW, Oh DH, Lee C, Yang SY. Allelic and haplotypic diversity of HLA-A, -B, -C, -DRB1, and -DQB1 genes in the Korean population. Tissue Antigens 2005;65:437-47.
5. Chung HY, Yoon JA, Han BY, Song EY, Park MH. Allelic and haplotypic diversity of HLA-A, -B, -C, and -DRB1 genes in Koreans defined by high-resolution DNA typing. Korean J Lab Med 2010;30:685-96.
6. Huh JY, Yi DY, Eo SH, Cho H, Park MH, Kang MS. HLA-A, -B and -DRB1 polymorphism in Koreans defined by sequence-based typing of 4128 cord blood units. Int J Immunogenet 2013;40:515-23.
7. Yoon JH, Shin S, Park MH, Song EY, Roh EY. HLA-A, -B, -DRB1 allele frequencies and haplotypic association from DNA typing data of 7096 Korean cord blood units. Tissue Antigens 2010;75:170-3.
8. Petersdorf EW, Gooley T, Malkki M, Horowitz M; International Histocompatibility Working Group in Hematopoietic Cell Transplantation. Clinical significance of donor-recipient HLA matching on survival after myeloablative hematopoietic cell transplantation from unrelated donors. Tissue Antigens 2007;69(S1):S25-30.
9. Lee SJ, Klein J, Haagenson M, Baxter-Lowe LA, Confer DL, Eapen M, et al. High-resolution donor-recipient HLA matching contributes to the success of unrelated donor marrow transplantation. Blood 2007;110:4576-83.
10. Tiercy JM. HLA-C incompatibilities in allogeneic unrelated hematopoietic stem cell transplantation. Front Immunol 2014;5:216.
11. Aubert O, Bories MC, Suberbielle C, Snanoudj R, Anglicheau D, Rabant M, et al., Risk of antibody-mediated rejection in kidney transplant recipients with anti-HLA-C donor-specific antibodies. Am J Transplant 2014;14:1439-45.
12. Willicombe M, Brookes P, Sergeant R, Santos-Nunez E, Steggar C, Galliford J, et al. De novo DQ donor-specific antibodies are associated with a significant risk of antibody-mediated rejection and transplant glomerulopathy. Transplantation 2012;94:172-7.
13. Deng Z, Wang D, Xu Y, Gao S, Zhou H, Yu Q, et al. HLA-C polymorphisms and PCR dropout in exons 2 and 3 of the Cw*0706 allele in sequence-based typing for unrelated Chinese marrow donors. Hum Immunol 2010;71:577-81.
14. Lee KW and Jung YA. Refined information on alleles belonging to the C*07:01/07:06/07:18 group in the Korean population. Human Immunol 2011;72:723-6.
15. The Comprehensive R Archive Network. http://cran.us.r-project.org/ (Updated on Oct 2014).
16. Nakajima F, Nakamura J, Yokota T. Analysis of HLA haplotypes in Japanese, using high resolution allele typing. MHC 2001;8:1-32.
17. Saito S, Ota S, Yamada E, Inoko H, Ota M. Allele frequencies and haplotypic associations defined by allelic DNA typing at HLA class I and class II loci in the Japanese population. Tissue Antigens 2000;56:522-9.
18. Trachtenberg E, Vinson M, Hayes E, Hsu YM, Houtchens K, Erlich H, et al. HLA class I (A, B, C) and class II (DRB1, DQA1, DQB1, DPB1) alleles and haplotypes in the Han from southern China. Tissue Antigens 2007;70:455-63.
19. Mack SJ, Tu B, Lazaro A, Yang R, Lancaster AK, Cao K, et al. HLA-A, -B, -C, and -DRB1 allele and haplotype frequencies distinguish Eastern European Americans from the general European American population. Tissue Antigens 2009;73:17-32.
20. Dunne C, Crowley J, Hagan R, Rooney G, Lawlor E. HLA-A, B, Cw, DRB1, DQB1 and DPB1 alleles and haplotypes in the genetically homogenous Irish population. Int J Immunogenet 2008;35:295-302.
21. Ruggeri L, Mancusi A, Burchielli E, Aversa F, Martelli MF, Velardi A. Natural killer cell alloreactivity in allogeneic hematopoietic transplantation. Curr Opin Oncol 2007;19:142-7.
22. Roh EY, In JW, Shin S, Yoon JH, Park KU, Song EY. Performance of LIFECODES HLA-DQB1 typing kit using Luminex platform in Koreans. Ann Lab Med 2015;35:123-7.