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HLA-linked Genetic Markers in Koreans

Joo Deuk Kim, In Hong Choi and Se Jong Kim

Department of Microbiology, Yonsei University College of Medicine, Seoul, Korea

The polymorphic variants of HLA-linked genetic markers GLO, Bf, C2 and C4 were determined in Koreans. The GLO^2 allele was found at a similar frequency as compared with other orientals and at higher frequency than in Caucasians. The gene frequencies of BfS and BfF showed different figures from those in other orientals and were simliar to Caucasians. The C2C allele was the highest variant as in other populations. The rare variant C2A was not observed in this study. The common variants of C4A alleles are C4A*3 and C4A*4. Among the C4B variants, C4B*1, C4B*2 and C4B* QO are common in that order. Several undefined electrophoretic variants C4A and C4B were observed in this study.

These findings suggest that the frequencies of various HLA-linked genetic markers can be used in anthropological studies.

Key Words : HLA, Immunogenetics, Glyoxalase I, Properdin factor B, C2, C4, Korean.

The structural genetic loci for three serum complement components, factor B(Bf), C2 and C4 (C4A or Rodgers and C4B or Chido) are very closely linked each other and to the loci of the major histocompatibility complex (MHC) on the short arm of the sixth chromosome of the human and they are very close to HLA-D/DR perhaps located between HLA-D/DR and HLA-B (Raum *et al.*, 1981). A genetic loci for the red cell glyoxalase I (GLO), also known as lactoyl-glutathione lyase, is linked to the MHC in the human, GLO catalyzes the irreversible conversion of glutathione and methylglyoxal to S-lactoyl-glutathione. GLO is structurally polymorphic and is found in both red and white cells, but red cells are a more convenient source of the enzyme for genetic studies (Valentine and Tanaka, 1961). Bf acts as the C3-proactivator in the alternate pathway of complement activation. Two common Bf alleles, F and S, and two rare alleles have been identified (Alper *et al.*, 1972). BfS and BfF have equal functional activity on the alternate pathway mediated hemolysis (Rittner and Bertrams, 1981). The genetically determined polymorphism of human C2 by a functional assay after the isoelectrofocusing of plasma was described (Alper, 1976; Hobart and Lachman, 1976). C2 was localized to the HLA through

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the study of families with cases of homozygous C2 deficiency (Fu *et al.*, 1974; Wolski *et al.*, 1975). Three alleles of C2, one a common variant C2C and two rare variants C2B and C2A were distinguished (Alper, 1976).

Of the HLA-linked complement components, C4 exhibits the greatest degree of polymorphism. There being two loci, the locus controlling acidic Rodgers positive variant is designated C4A (C4F) and that controlling the basic Chido positive variants is designated C4B (C4S). Six structural variants (C4A*1-6) and one null variant (C4A*QO) at C4A locus were distinguished. At C4B locus, there are three common structural variants and one null variant designated as C4B*1-3 and C4B*QO respectively.

These have been established that not only certain HLA antigens but also HLA-linked genetic markers; GLO, Bf, C2 and C4 alleles occur more frequently in patients than in healthy control populations, whereas other such genes may be reduced in frequency among patients. An association between BfFl and insulin dependent diabetes mellitus in Caucasians (Barbosa *et al.*, 1979; Kirk *et al.*, 1979; Raum *et al.*, 1979) and the demonstration that the rare C4 haplotype C4A*4:C4B*2 occurs together with the rare C2*B allele in 21-hydroxylase deficiency in Italian families (O'Neill *et al.*, 1982) were described recently. C2 deficiency (Mahowald *et al.*, 1979) and C4 deficiency (Awdeh *et al.*, 1981; Ballow *et al.*, 1979) have been reported to be associated with HLA.

It has been well known that the pattern of antigen frequencies of HLA antigens could be different in different ethnic groups. Since genetic polymorphisms of HLA-linked genetic markers have been demonstrated, the ethnic differences in the frequencies of HLA-linked gene alleles may also be expected. Therefore, the present study was attempted to perform the typing of four HLA-linked genetic markers (GLO, Bf, C2 and C4) in Koreans and to compare with other ethnic groups.

MATERIALS AND METHODS

Samples : Blood samples from 123 healthy unrelated Koreans in Seoul were collected in test tubes containing EDTA or ACD. Plasma was separated by centrifugation and stored at $-70^{\circ}C$ and thawed just before analysis.

Bf typing : Bf typing was performed by electrophoresis of plamsa in 1% agarose gel and the patterns were developed by immunofixation with anti-human Bf antiserum (Alper *et al.*, 1972).

GLO typing : GLO typing was carried out by starch gel electrophoresis of red cell lysates as described by Kömpf *et al* (1975).

C2 typing : Plasma samples were subjected to isoelectrofocusing in polyacrylamide gels followed by C2 specific hemolytic agarose overlay (Alper, 1976).

C4 typing : Typing of C4 electrophoretic variants was performed by electrophoresis of EDTA plasma treated with neuraminidase followed by overlaying anti-human C4 antiserum over the agarose and allowing it to react before washing in saline and staining with Coomassie brilliant blue (Awdeh and Alper, 1980).

Complement genetic nomenclature : The nomenclature for genetic polymorphism of Bf, C2 and C4 is that proposed earlier (Awdeh and Alper, 1980), which was designed to conform to the International System for Human Gene Nomenclature, 1979 (Shows *et al.*, 1979).

Statistical analysis : Gene frequencies for the different alleles of the individual loci determined by gene counting.

RESULTS AND DISCUSSION

The gene frequencies of GLO alleles are given in Table 1. The GLO^2 allele (gf=0.910) is the highest allele followed by GLO^1 allele (gf=

Table 1. GLO gene frequencies in Koreans and other populations

Allele	Koreans (N=78)	Chinese[a] (N=140)	Japanese[a] (N=50)	Caucasians[a] (N=70)
GLO^1	0.090	0.133	0.046	0.431
GLO^2	0.910	0.865	0.954	0.569

a Miniter et al. (1981)

Table 2. Bf gene frequencies in Koreans and other populations

Allele	Koreans (N=122)	Chinese[a] (N=140)	Japanese[a] (N=50)	Caucasians[a] (N=780)
S	0.783	0.892	0.909	0.742
F	0.217	0.104	0.091	0.225
S1	N.O.	0.004	N.O.	0.017
F1	N.O.	N.O.	N.O.	.0.016

a Miniter et al. (1981)

N.O. : Not observed

0.090). This is quite similar to the frequencies observed in Chineses and Japanese (Miniter *et al.*, 1981). However, our study shows a higher frequency of GLO^2 than that seen in Caucasians (O'Neill *et al.*, 1979).

The gene frequencies of Bf alleles in Koreans compared with other populations are shown in Table 2. The most common Bf allele was BfS (gf=0.783), followed by the BfF (gf=0.217). When comparing the reported frequencies of Bf alleles in other Oriental populations (Miniter *et al.*, 1981), some differences can be seen. In Blacks (Budowle *et al.*, 1981; Klouda et al., 1983), the ratio of BfS allele to BfF allele was reversed as compared not only with Koreans in our study but also with other orientals and Caucasians. Neither of the rare variant of Bf, BfSl or BfFl was observed in this study. There is great variation in the frequency of the rare Bf variants between different ethnic groups. A high frequency of BfFl in Basques (Ohayon *et al.*, 1980) and in the Spanish population

Table 3. C2 gene frequencies in Koreans and other populations

Allele	Koreans (N=60)	Chinese[a] (N=160)	Japanese[a] (N=50)	Caucasians[a] (N=780)
C	0.992	0.949	1.00	0.954
B	0.008	0.039	N.O.	0.046
A	N.O.	0.001	N.O	N.O

a Miniter et al. (1981)

N.O. : Not observed

Table 4. C4A and C4B gene frequencies in Koreans and other populations

Allele	Koreans (N=82)	Italian[a] (N=38)
C4A*QO	0.061	0.1842
1	N.O.	0.0000
2	N.O.	0.1184
3	0.701	0.6184
4	0.226	0.0789
5	N.O.	0.0000
6	N.O	0.0000
C4B*QO	0.116	0.1316
1	0.543	0.1316
2	0.287	0.2368
3	N.O.	0.0000

a O'Neill et al. (1982)

QO : Null gene

N.O. : Not observed

(Rodriguez-Cordoba *et al.*, 1981) was reported. Furthermore, the BfFl has not been reported in other Orientals (Horai *et al.*, 1979).

Table 3 demonstrates the gene frequencies of C2 allele in Koreans. The C2C allele occurred at a gene frequency of 0.992 in Koreans. The rare variant, C2B occurred at a gene frequency of 0.008 and no C2A variant was observed in this study. Koreans, other orientals (Miniter *et al.*, 1981; Tokunaga *et al.*, 1981) and Caucasians (Miniter *et al.*, 1981; O'Neill *et al.*, 1979) show similar frequencies of C2C and

C2B alleles.

The gene frequencies for the allele of the C4A and C4B loci in Koreans are summarized in Table 4. The most commonly observed variant of C4A in Koreans was, C4A*3 (gf= 0.701), followed by C4A*4(gf=0.226). A lower frequencies of C4A*QO and higher C4A*4 variant were observed in Koreans than in Italians (O'Neill *et al.*, 1982). The C4A*2, a relatively common variant in the Italian population, was not observed in this study. The common variants of C4B locus were C4B*1 (gf=0.543), C4B*2 (gf=0.287) and C4B*QO(gf=0.116) . These gene frequencies in Koreans are very similar to that in the Italian population. Several rare variants which showed different electrophoretic patterns from C4A*1-6, C4A*QO, C4B*1-3 and C4B*QO were observed at low frequencies in this study.

The reliable detection of polymorphism of HLA-linked genetic markers, especially highly polymorphic C4 as demonstrated in this study and others, suggests that it could prove to be a very valuable genetic marker not only in anthropological studies but also disease association studies.

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