Letters to the Editor

Increased risk of human leukocyte antigen-G gene variants in Behçet's disease

Sirs,

Behçet's disease (BD, MIM 109650) is a multisystemic chronic inflammatory disease characterized by the recurrence of a variety of symptoms. Immunological abnormalities and variants of inflammatory molecules also appear to be involved in the etiology of BD.

Human leukocyte antigen-G (HLA-G, MIM 142871) is a nonclassical MHC class Ib molecule, which is expressed during pathological processes, viral infections, and chronic cutaneous inflammatory diseases such as psoriasis, atopic dermatitis, and ulcerative colitis in peripheral tissues (1, 2). But it is not likely to be expressed in the skin of healthy individuals. HLA-G molecules function as tissue-protective molecules during inflammatory responses and exert inhibitory effects on immune cells. They act as a shield against inflammatory aggression as well (3, 4). HLA-G is involved in a wide variety of biological processes including the modulation of immune response, inhibition of NK/T-mediated cytotoxicity, and the regulation of cytokine secretion in Th1 and Th2 cells (5). Polymorphisms in *HLA-G* gene may influence the expression or function of the HLA-G molecule. *HLA-G* polymorphisms and haplotypes, which affect its isoforms and protein levels, may be associated with the susceptibility to BD.

Two hundred and eighty eight BD patients (male: 145, female: 143), age range: 16-66 years at diagnosis) were diagnosed and classified according to the proposed criteria by both International Study Group for Behcet's disease and BD Research Committee of Japan (6). Two hundred and sixty six unrelated healthy controls were also subjected for the study. All subjects enrolled in this study had Korean ethnicity.

Four polymorphisms of *HLA-G -727C>T* in the promoter, *1074A>T* (Thr31Ser) in the a1 domain, *1597delC* (Leu130frameshift) in the a2 domain, and *3775G>C* of the 3'-UTR in exon 8 were analyzed by the PCR-RFLP (7-8), and *3741_3754ins14* in exon 8 was detected by the PCR (9).

All genotype distributions of the *HLA-G -727C>T*, *1074A>T*, *1597delC*, *3741_-3754ins14*, and *3775G>C* polymorphisms for the BD patients and the controls satisfied the Hardy-Weinberg equilibrium. The frequency of the *3741_3754*ins14* allele (p = 0.035, OR [odds ratio] = 1.5, 95% CI [confidence interval] = 1.03-2.24) was significantly higher in the BD patients than in the controls by the Chi-square test. Those with the *3741_3754*ins14/*ins14* homozygote were found to have twice the risk of BD than the controls. Although being not significant, the trend toward an increased frequency of the *HLA-G 1597*delC* allele was more prevalent in the BD patients than in the controls (0.042 *vs*. 0.024). With regard to genotype or allele frequencies, the *HLA-G -727C>T* and *3775G>C* were similar between the BD patients and the controls. The *1074A>T* (Thr31Ser) variant in the α 1 domain was detected in neither the BD patients nor the controls among Koreans (Table I). Seven haplotypes were observed in our Korean sample by using the PHASE program. In a case-control permutation test, the frequency of haplotypes was not found to differ between the total BD patients and the controls (p = 0.240), but the

Table I. Genotype, allele, and haplotype frequencies of the *HLA-G* in BD patients and in controls.

HLA-G Region		BD n = 288 (%)	Controls n = 266 (%)	p	OR (95%CI)
Promoter					
-727C>T	*-727*C/*C*	282 (97.9)	262 (98.5)		
	*-727*C/*T*	6 (2.1)	4 (1.5)		
	*-727*T/*T*	0 (0.0)	0 (0.0)		
	*-727*T*	0.010	0.008		
Exon 2					
1074A>T					
	*1074*A /*A*	288 (100.0)	266 (100.0)		
Thr31Ser	*1074*A /*T*	0 (0.0)	0 (0.0)		
	*1074*T/*T*	0 (0.0)	0 (0.0)		
	1074 T*	0	0		
Exon 3					
1597delC					
	*1597*C/*C*	265 (92.0)	253(95.1)		
Leu130framshift	*1597*C/*delC*	22 (7.6)	13 (4.9)		
	*1597*delC/*delC*	1 (0.4)	0 (0.0)		
	*1597*delC*	0.042	0.024		
Exon 8					
3741_3754ins14	*3741_3754*del14/*del14*	147 (51.0)	163 (61.3)	0.015	0.7 (0.47-0.92)
	*3741_3754*del14/*ins14*	116 (40.3)	91 (34.2)		
	*3741_3754*ins14/*ins14*	25 (8.7)	12 (4.5)	0.050	2.0 (0.99-4.09)
	*3741_3754*ins14*	0.288	0.212	0.035	1.5 (1.03-2.24)
3775G>C	*3775*G/*G*	50 (17.4)	50 (18.8)		
	3775*G/*C	144 (50.0)	137 (51.5)		
	*3775*C/*C*	94 (32.6)	79 (29.7)		
	*3775*C*	0.576	0.555		
Haplotype					
-727C 1074A 1597C 3741_3754del14 3775G		0.408	0.436		
-727C 1074A 1597C 3741_3754del14 3775C		0.301	0.346		
-727C 1074A 1597C 3741_3754ins14 3775C		0.231	0.183	0.046	1.3 (1.00-1.81)
-727C 1074A 1597delC 3741_3754ins14 3775C		0.035	0.019		
-727C 1074A 1597C 3741_3754ins14 3775G		0.015	0.009		
-727T 1074A 1597delC 3741_3754ins14 3775C		0.007	0.005		
-727T 1074A 1597C 3741_3754del14 3775C		0.003	0.002		

haplotypic frequency of *HLA-G -727C 1074A 1597C 3741_3754ins14 3775C* was higher in the BD patients (p = 0.046) than in the controls (Table I).

HLA-G 3741_3754ins14 variant leads to splice out the 92-bp of exon 8 in 3'-UTR and induces a significantly lower mRNA level and a lower concentration of soluble HLA-G than the corresponding HLA-G mRNA isoform with the deleted 14bp (10).

In conclusion, the *HLA-G 3741_3754ins14* variant and the haplotype carrying this variant are associated with an increased risk for BD. These results suggest that the presence of the HLA-G *3741_3754ins14* variant which is related with lower mRNA level is probably associated with the development of BD.

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A molecular basis for the absence of familial Mediterranean fever in Ethiopian Jews

Sirs,

Familial Mediterranean fever (FMF) is the most prevalent hereditary inflammatory disorder (1). Four ethnic groups are classically affected: Armenians, Arabs, Turks and Jews. However, FMF is not equally shared among Jews. Non Ashkenazi Jews, i.e. Sefardi Jews, are far more affected than Jews from Europe.

Ethiopian Jews have been arriving in Israel in ever increasing numbers since around 1979. Today more than 50,000 Ethiopian Jews are living in Israel (2). In our experience, as well as in the English literature, no case of FMF has been reported in this community

In the present study, we have searched for MEFV mutations (3, 4) in 95 unrelated unaffected Ethiopian Jews.

Ninety five adult Jewish subjects from Ethiopian ancestry (44 males, 51 females) were enrolled. All of them arrived in Israel during the last five years and were living in two immigration centers in the north of Israel. There is no identified Christian Ethiopian community in Israel so we could not include a control group of "non Jewish Ethiopian population" in our study. The subjects had no special medical history except for infections: one active pulmonary tuberculosis, one hepatitis B infection with cirrhosis and one HIV positive. The study received Helsinki committee approval and informed consent in Hebrew and Ethiopian language was obtained from every participant.

No subject was found to be either homozygous or compound heterozygous at the MEFV locus. One mutated allele was identified in 14 individuals. We found none of the most penetrant/severe mutations associated with FMF, i.e. at codons 694 and 680, nor other mutations in exon 10. A common polymorphism was identified, however (P706, 1%). In contrast, we detected a relatively high rate of mutations E148Q (6%), and P369S (1%) (Table I).

Our study suggests a molecular basis for the absence of FMF in Ethiopian Jews.

The present report may contribute to the persistent debate of whether E148Q is a polymorphism or a true mutation (5-7). Recent studies have suggested that this mutation/polymorphism may enhance inflammation non-specifically, of possible advantage during evolution (8).

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Table I. MEFV sequence variants in 95 unaffected Ethiopian Jews.

	N	%	95%CI
E148Q	11	6%	0.03-0.09
P369S	2	1%	0.001-0.03
P706*	1	0.5%	0.001-0.02
Mutations in exon 10	0	0%	0-0.02
All mutations*	14	7.5%	0.04-0.11
Carrier estimation		14%	0.09-0.19
Genetic FMF estimation		0.5%	0.001-0.02

* P706 is considered a polymorphism