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석사학위청구논문

Association between *CR4*(*CD11c /CD18*) Gene Polymorphisms and Behcet's Disease

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성신여자대학교 대학원

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이 논문을 석사학위논문으로 제출함

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성신여자대학교 대학원

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인 준 서

박수림의 석사학위 논문으로 인준함.

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논 문 개 요

보체 수용체 4 (Complement receptor 4; CR4 (CD11c/CD18)는 β2-인테그린 으로, 호중구, 자연살해세포, 수지상, 대식세포와 같은 염증관여 면역 세포에 발현하여 염증 부위로 이동시킨다. β2-인테그린의 발현 증가는 면역 세포의 이동과 싸이토카인 생산을 조절함으로써 베체트병 (Behcet's disease, BD)과 같은 염증성질환의 염증반응에 중요한 역할을 한다. 이 논문은 베체트 병 환자 305 명과 건강인 266 명의 *CR4* (*CD11c/CD18*) 유전자 5' near gene, 엑손, 인트론, 3'UTR, 6 곳의 단일염기다형성(SNPs)을 PCR-RFLP 방법을 사용하여 베체트병에서 *CR4* 유전자의 유전적 연관성을 분석 하였다.

*CD11c*와 *CD18* 각SNP들의 유전형과 대립인자 빈도는 건강인과 BD환자군에서 모두 하디-와인 버그 평형에 통계적으로 어긋나지 않았다. *CD18* 의 rs235326과 rs684 두 SNPs간의 강한 연관을 건강인과 BD환자군에서 보였으며 (각각 D' = 0.997, r^2= 0.038; D' = 0.806, r^2= 0.017), *CD11c* rs2230429 *C>G*, rs2929*G>A* 와 *CD18* rs2070946*T>C,* rs760456*C>G* SNPs간의 연관은 나타나지 않았다.

CD11c rs2230429 (exon15) *C/C* 유전형, *C* 대립인자와 rs2929 (3'UTR) *G/G* 유전자형, *G* 대립인자 의 빈도는 각각 p = 0.0004, OR = 1.8, 95% CI = 1.31 - 2.56; p = 0.007, OR = 1.7, 95% CI = 1.15 - 2.47; p = 0.021, OR = 1.8, 95% CI = 1.06 - 2.05; p = 0.013, OR = 1.4, 95% CI = 1.07 - 1.78 로 BD환자군이 건강인군보다 통계적으로 유의하게 높았다. *CD18* rs235326 (exon11) *C/C* 유전형과 *C* 대립인자의 빈도는 각각 p = 0.002, OR = 1.7, 95% CI = 1.20 - 2.36; p = 0.022, OR = 1.6, 95% CI = 1.07 - 2.36 로 BD환자군이 건강인보다 통계적으로 유의하게 높았다. BD환자군 내에서 임상적 증상을 보이는 사람과 보이지 않는 사람간의 비교를 해 본 결과,

흥미롭게도 *CD11c* rs2929 *C/C* 유전형빈도는 관절염 증상을 보이는 사람이 보이지 않는 사람보다 유의하게 높았고 (p = 0.012), 신경계 병변을 보이는 사람이 보이지 않는 사람보다 유의하게 낮았다 (p = 0.012).

CD11c rs2230429 *C/C* 와 *CD18* rs235326 *C/C* 유전형을 가진 BD 환자군과 *CD11c* rs2230429 *C/C,* rs2929 *G/G* 와 *CD18* rs235326 *C/C* 유전형을 가진 BD 환자군의 빈도는 각각 p = 0.003, OR = 1.7, 95% CI = 1.19 - 2.40; p = 0.005, OR = 1.9, 95% CI = 1.20 - 2.90 로 건강인군에 비해 통계적으로 유의하게 높았다. *CD11c* rs2230429 *C/C* 과 rs2929 *G/G* 과 *CD18* rs235326 *C/C* 을 모두 가진 BD 환자군의 빈도는 건강인군에 비해 통계적으로 유의하게 높은 것으로 나타났다. *CD11c* 의 일배체형 rs2230429C-rs2929G 빈도는 BD 환자군이 건강인군에 비해 통계적으로 유의하게 높았고 (p = 0.0002, OR = 1.5, 95% CI = 1.23 - 1.97), *CD11c* 의 일배체형 rs2230429G - rs2929G 와 *CD11c* rs2230429*G* - rs2929*A* 빈도는 BD 환자군이 건강인군에 비해 통계적으로 유의하게 낮았다 (각각 p = 0.044, OR = 0.7, 95% CI = 0.53 - 0.99; p = 0.0009, OR = 0.5, 95% CI = 0.31 - 0.74). *CD18* 의 일배체형 rs2070946*A* - rs235326*C* - rs760456*G* - rs684*G* 빈도는 BD 환자군이 건강인군에 비해 통계적으로 유의하게 높았고 (p = 0.007, OR = 1.5, 95% CI = 1.19 - 1.91), *CD18* 의 일배체형 rs2070946*A* - rs235326*T* - rs760456*C* - rs684*G* 빈도는 BD 환자군이 건강인군에 비해 통계적으로 유의하게 낮았다 (p = 0.003, OR = 0.4, 95% CI = 0.26 - 0.77). 따라서 *CD11c* rs2230429*C>G*, rs2929*G>A* 와 *CD18* rs235326*T>C* 은 베체트병 발병과 positive 연관이 있으며, *CD11c* Pro517Arg, 3'UTR 과 *CD18* Val441Val 의 유전자 변이는 BD 발병위험인자의 하나로써 활용할 수 있을 것이다.

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Introduction

Behcet's disease (BD) is a systemic inflammatory disorder, characterized by four major symptoms consisting of oral ulcerations, ocular lesions, skin lesions and genital ulcerations and other manifestations, including the vascular system, central nervous system, gastrointestinal tract, lungs, kidneys, and joints (Zierhut et al., 2003). The pathogenesis of BD is still uncertain, but the causes of BD are a complex interaction of genetic and environmental factors and immunological dysfunction such with phenotypic and functional lymphocyte abnormalities, hyperactivity of neutrophils, over expression of several pro-inflammatory and T-helper-1 (Th1) cytokine (Karasneh et al., 2005; Krause et al., 2008; van et al., 2009; Mendes et al., 2009).

The leukocytes and leukocyte migration into inflammatory site by the interaction between adhesion molecules and integrins on endothelia cell play an important role in the development of inflammatory diseases such as BD (Efthimiou et al., 1989; Sahin et al., 1996). The neutrophils and monocytes increase motility and enhance adhesion in endothelial cells by the increased expression of β2-integrin lymphocyte function-associated antigen-1 (LFA-1, CD11a/ CD18) and Intercellular adhesion molecule-1 (ICAM-1) in BD inpatients (Sahin et al., 1996; Zierhut et al., 2003; Davtyan et al., 2006). Also, *ICAM-1* mutation is associated with BD susceptibility in Korean patients (Kim et al., 2003), as well as the relation of LFA-1 and macrophage antigen complex -1 (Mac-1), which is a member of β2-integrins with BD and has been discussed before (Sahin et al.,

1996). However, any other β2-integrins complement receptor 4(CR4, CD11c/CD18) and BD association has not been described until now.

The β2-integrins (CD11/CD18) family is a heterodimer that is comprised of β2 chain (CD18) with one of four α chains and termed $\alpha_L\beta_2$ (lymphocyte function-associated antigen-1; LFA-1), $\alpha_M\beta_2$ (macrophage-1 antigen; Mac-1, CR3), $\alpha_X\beta_2$ (CD11c/CD18, p150/95, CR4) and $\alpha_D\beta_2$ (CD11d/CD18). β2-integrins are best known for cell-cell adhesion, tissue-specific homing, leukocyte activation and trafficking during inflammation in immune responses and phagocytosis of complement opsonized particles (Gahmber et al., 1997; Harris et al., 2000; Oreshkova et al., 2009).

CR4(CD11c/CD18) is expressed on monocytes/macrophages, neutrophils, dendritic cells, NK cells and lymphocytes and can bind to fibrinogen, iC3b, the endothelial ligand ICAM-1, ICAM-2, ICAM-4 and vascular cell adhesion molecule-1(VCAM-1) (Postigo et al., 1991; Bullard et al., 2007; Sadhu et al., 2007; Rosas et al., 2009; Wu et al., 2009; Gower et al., 2011). The functional role of CR4 is not clearly known yet. But, it appears to be similar to those of other β2-integrins member Mac-1(CD11b/CD18). CR4(CD11c/CD18) along with Mac-1(CD11b/CD18) are important in complement-mediate phagocytosis, and their up-regulation in neutrophil activation is closely coordinated (Rezzonico et al., 2001; Hellmig et al., 2005; Hu et al., 2010). And the binding of fibrinogen to CR4(CD11c/CD18) and Mac-1(CD11b/CD18) on human mononuclear phagocytes results in nuclear factor- κB(NF-κB) activation and enhanced interleukin (IL)-8 production (Sitrin et al.,1998). Also, the synthesis of IL-8, macrophage inflammatory protein-1α (MIP-1α) and MIP-1β is stimulated by triggering CD11b and

CD11c α chains of β2-integrins on primary human monocytes (Rezzonico et al., 2001). The CD11c/CD18 triggers to IL8 gene activation in polymorphonuclear neutrophils during the inflammatory response on ligand binding (Walzog et al., 1999). The IL-8 and MIP-1α are known to play an important role in inflammatory response of BD (Ozer et al., 2005; Mendes et al., 2009). The other investigators suggested that CD11c on the dendritic cells and macrophages may function at antigen-presentation step by binding to ICAM-1 on T cells. Also, engagement of CD11c may lead to cytokine production by the antigen presenting cell (APC) (Meunier et al., 1994; Sadhu et al., 2007). Moreover, CD11c/CD18 plays an important role in leukocyte migration into inflammatory site (Bullard et al., 2007). Also, CD11c/CD18 is known to be involved in primary stage of chemokine-stimulated adhesion and migration of cell on fibrinogen (Kukhtina et al., 2011).

CR4 (CD11c/CD18) obtains high affinity to endothelial ligands by extensive conformation changes in both α and β subunits (Alon., 2010). *CD11c* and *CD18* gene mutation may be an effect of conformation change of CR4; thereby, CR4 (CD11c/CD18) may be involved in inflammatory response such as increased motility of leukocyte and cytokine production. Whether CD11c/CD18 affects inflammation response of BD has not been demonstrated yet, but CD11c/CD18 may be involved in inflammation events for BD by enhancing inflammatory cytokines production and leukocytes migration.

This study investigated whether *CR4*(*CD11c/CD18*) gene polymorphisms is associated with BD patients.

Subjects and Methods

Subjects

A total of 305 Behcet's disease patients (148 males and 157 females) and 266 healthy controls in Korea were included in this case-control study. Behcet's disease patients were recruited from the Ajou University of Medicine, Suwon Korea and Behcet's Disease Speciality Clinic of Severance Hospital Yonsei University College of Medicine, Seoul, Korea. BD fulfilled the clinical criteria proposed by the International Study Group for Behçet‘s Disease (1990). Consents were obtained from subjects, and the Institutional Review Board approved protocols of this study. The distribution of clinical features in patients with BD is presented in the Table 1.

Genotyping

Genomic DNA was extracted from peripheral blood using a QIAamp Blood kit (Quiagen, Hilden, Germany).

The National Center for Biothechnology Information (NCBI) was used to identify SNPs. Genotyping of the *CD11c* rs2230429 (*50C>G*), *CD11c* rs2929 (*80G>A*), *CD18* rs235326 (*1323T>C*), *CD18* rs760456 (*784C>G*), *CD18* rs684 (*122G>A*), and *CD18* rs2070946 (*-4+7372A>G*) were determined by the polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) method. For detection of the polymorphism, 10 ㎕ reaction volume containing 15 ng of DNA was amplified with 10 mM Tris (pH 8.0), 40

mM KCl, 1.5 mM $MgCl_2$, 200 M dNTP, 5 pmol each primer and 0.38 unit Taq polymerase (Bioneer, Korea) on GeneAmpPCR system 9700 (Applied Biosystems, Foster City, CA, USA).

The *CD11c* rs2230429 *C>G* polymorphism was amplified using sense 5'-TGGAGAAGGTGGTGGTGTGAT-3' and antisense 5'-TCCCAAGACTCCGTGAAAC AG-3' primers which were digested with the restriction enzyme *MspAI* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed by 32 cycles of 30s at 94 C, 45s at 63 C, 1 min at 72 C, and a final extension at 72 C for 5 min. The *CD11c* rs2929 *G>A* polymorphism was amplified using sense 5'-AGAAAACGGGACACAGACCC-3' and antisense 5'-TCTCCCAAAGACAGGCA-3' primers which were digested with the restriction enzyme *NlaIV* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed by 32 cycles of 30s at 94 C, 45s at 62 C, 1 min at 72 C, and a final extension at 72 C for 5 min. The *CD18* rs2070946 *A>G* polymorphism was amplified using sense 5'-TATCCTCCTTGGCTGGTTCC-3' and antisense 5'-ATTCACACTTGGGAGAGGGG-3' primers which were digested with the restriction enzyme *BsoBI* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed by 32 cycles of 30s at 94 C, 45s at 62 C, 1 min at 72 C, and a final extension at 72 C for 5 min. The *CD18* rs235326 *T>C* polymorphism was amplified using sense 5'-CGGCCACAGAGTGCATCCAGGA-3' and antisense 5'-GCCGCACTCCAAGAAGCC CTTG-3' primers which were digested with the restriction enzyme *AvaII* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed

by 36 cycles of 30s at 94 C, 45s at 62 C, 1 min at 72 C, and a final extension at 72 C for 5 min. The *CD18* rs760456 *C>G* polymorphism was amplified using sense 5'-GGCTCAGAATCTCCTCACTCA-3' and antisense 5'-TCCAGACAAGACCCCCGAG GG-3' primers which were digested with the restriction enzyme *Tsp45I* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed by 30 cycles of 30s a t 94 C, 40s at 64 C, 1 min at 72 C, and a final extension at 72 C for 5 min. The rs684 *G>A* polymorphism was amplified using sense 5'-CACCAGGATAATCCCCTTTTCA-3' and antisense 5'-TTGGCACCACCTTTAATC AGAC-3' primers which were digested with the restriction enzyme *AciI* (New England Biolabs, USA). The PCR conditions were initial denaturation of 5 min at 94 C followed by 32 cycles of 30s at 94 C, 45s at 62 C, 1 min at 72 C, and a final extension at 72 C for 5 min. Digested PCR products were electrophoresed in 8% polyacrylamide gel and stained with ethidium bromide in order to be seen under UV.

Statistical Analysis

The differences of allele frequencies and genotype distribution between Behcet's disease patients and controls were examined by the χ^2 test using SAS v.9.1.3 (SAS Institute, Cary, NC). Odds ratios (OR) with 95% CI were obtained. The *p*-values <0.05 were regarded as statistically significant. For multiple comparison analysis by using the version 4.2 of Haploview program (http://www.broadinstitute.org/haploview/haploview) has been subjected permutation correction: the Permutation *p*-values were obtained by running 1000 per mutations (permutation corrected *p*-vaule = as significant level). The Hardy-

Weinberg equilibrium and linkage disequilibrium (LD) were analyzed using the R program v.2.12.2 (http://www.r-projects.org/). LD ; the most frequently used is D' > 0.8 (Carlson et al., 2004). The version 4.2 of Haploview program (http://www.broadinstitute.org/haploview/haploview) was used to infer haplotypes.

Results

The genotype and allele distributions of the *CD11c* and *CD18* polymorphisms in Behcet's disease (BD) patients and healthy controls are shown in Table 2. Each genotype and allele frequency of the *CD11c* and *CD18* polymorphisms in BD patients and healthy controls were in the Hardy-Weinberg equilibrium. The homozygous *CD11c* rs2230429 *C/C* genotype (p = 0.0004, OR = 1.8, 95% CI = 1.31 - 2.56) and *C* allele frequencies (p <.0001, OR = 1.7, 95% CI = 1.31 - 2.26, permutation p = 0.001) were significantly higher in BD patients than in healthy controls. The homozygous *CD11c* rs2230429 *G/G* (Pro517Arg) genotype was significantly lower in BD patients than in healthy controls (p = 0.021, OR = 0.4, 95% CI = 0.22 - 0.99). *CD11c* rs2929 *G/G* genotype was significantly higher in BD patients than in healthy controls (p = 0.021, OR = 1.5, 95% CI = 1.06 - 2.05) and *G* allele frequencies (p = 0.013, OR = 1.4, 95% CI = 1.07 – 1.78) were significantly higher in BD patients than in healthy controls. The homozygous *CD18* rs235326 *C/C* (Val441Val) genotype (p = 0.002, OR = 1.7, 95% CI = 1.20 - 2.36) and *C* allele frequencies (p = 0.001, OR = 1.6, 95% CI = 1.20 - 2.10, permutation p = 0.007) were significantly higher in BD patients than in healthy controls. There was no significant difference in the genotype frequency of the *CD18* rs2070946 *A>G*, rs760456 *C>G* and rs684 *G>A* (Table 2).

The linkage disequilibrium (LD) between 2SNPs of *CD11c* was not observed. However, between 2SNPs of *CD18* (rs235326 and rs684) were strong LD in control

and BD (D' = 0.997, r^2 = 0.038; D' = 0.806, r^2 = 0.017, respectively; Figure 1). The frequency of haplotype,*CD11c* rs2230429C - rs2929G, revealed increased susceptib ility to BD as compared with controls (p = 0.0002, OR = 1.5, 95% CI = 1.23 – 1.97, permutation p = 0.001) while, that of *CD11c* rs2230429G - rs2929G and *CD11c* rs2230429*G* - rs2929*A* revealed decreased susceptibility to BD as compared with controls (p = 0.044, OR = 0.7, 95% CI = 0.53 - 0.99; p = 0.0009, OR = 0.5, 95% CI = 0.31 - 0.74, permutation p = 0.004, respectively). The frequency of haplotype *CD18* rs2070946*A* - rs235326*C* - rs760456*G* - rs684*G* revealed increased susceptibi lity to BD as compared with controls (p = 0.007, OR = 1.5, 95% CI = 1.19 – 1.91) while, that of *CD18* rs2070946*A* - rs235326*T* - rs760456*C* - rs684*G* revealed decreased susceptibility to BD as compared with controls (p = 0.003, OR = 0.4, 95% CI = 0.26 - 0.77, permutation p = 0.012)(Table 6).

The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/C* and rs2929*G/G* was significantly higher compared with controls (36.0% vs. 22.9%; p = 0.0006). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/G* or *G/G* and rs2929 *G/A* or *A/A* was significantly lower compared with controls (18.4% vs. 29.7%; p <.0001; Figure 2). The frequency of BD patients who were carrying the *CD18* rs2070946*A/A* and rs235326 *C/C* was significantly higher compared with controls (56.0% vs. 43.2%; p = 0.002). The frequency of BD patients who were carrying the rs2070946 A/G or *G/G* and *CD18* rs2353226 *C/T* or *T/T* was significantly lower compared with controls (7.9% vs. 13.9%; p = 0.020; Figure 3). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/C* and

CD18 rs235326 *C/C* was significantly higher compared with controls (40.3% vs. 28.6%; p = 0.003). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/G* or *G/G* and *CD18* rs2353226 *C/T* or *T/T* was significantly lower compared with controls (10.5% vs. 25.9%; p <.0001; Figure 4). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/C* and rs2929 *G/G* and *CD18* rs235326 *C/C* was significantly higher compared with controls (22.6% vs. 13.6%; p = 0.005). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/G* or *G/G* and rs2929*G/G* and *CD18* rs235326 *C/T* or *T/T* was significantly lower compared with controls (4.9% vs. 10.5%; p = 0.011). The frequency of BD patients who were carrying the *CD11c* rs2230429 *C/G* or *G/G* and rs2929 *G/A* or *A/A* and *CD18* rs235326 *C/T* or *T/T* was significantly lower compared with controls (5.6% vs. 15.4%; p = 0.0001; Figure 5).

BD is a multisystemic inflammatory disease, and patients usually showed three or more symptoms. This study analyzed the relationship between the SNPs and various clinical manifestations such as oral, genital ulcers, ocular lesions, arthritis, vascular involvement, gastrointestinal involvement and neurologic involvement. The frequency of major genotype *CD11c* rs2230429 *C/C* that was observed was significantly higher in BD patients with clinical features of major symptoms (oral ulcers: p = 0.0005, OR = 1.8; skin lesions: p = 0.0004, OR = 1.9; genital ulcers: p = 0.0002, OR = 2.0; ocular lesions: p = 0.0008, OR = 1.9) The frequency of major genotype *CD11c* rs2929 *G/G* that was observed was significantly higher in BD patients with clinical features of major symptoms (oral ulcers: p = 0.030, OR = 1.4; skin lesions: p = 0.024, OR = 1.5; genital

ulcers: p = 0.034, OR = 1.5; ocular lesions: p = 0.045, OR = 1.5) and in two of minor symptoms (arthritis: p = 0.0003, OR = 2.4; vascular: p = 0.043, OR = 2.1) in BD patients than in healthy controls (Table 3). The frequency of major genotype *CD18* rs235326 *C/C* that was observed was significantly higher in BD patients with clinical features of major symptoms (oral ulcers: p = 0.004, OR = 1.6; skin lesions: p = 0.005,OR = 1.6; genital ulcers: p = 0.0003, OR = 1.9; ocular lesions: p = 0.015, OR = 1.6) and in two of minor symptoms (arthritis: p = 0.009,OR = 1.9; neurologic involvement: p = 0.025, OR = 7.6) in BD patients than in healthy controls (Table 4).

Interestingly, the distribution of major genotype *CD11c* rs2929 *G/G* was observed to be significantly higher in BD patients with arthritis than in BD patients without arthritis (p = 0.012). And, although the number of BD patients with neurologic involvement is small, but the distribution of major genotype *CD11c* rs2929 *G/G* was observed to be significantly higher in BD patients with neurologic involvement than in BD patients without neurologic involvement (p = 0.012) (Table 5). Also, the frequency of haplotype *CD11c* rs2230429G - rs2929G revealed susceptibility to be higher in BD patients with arthritis than in BD patients without arthritis (p = 0.005, OR = 1.9, 95% CI = 1.21 – 3.00, permutation p = 0.016, Table 7).

Discussion

Complement receptor 4 (CR4, CD11c/CD18) is heterodimer that is comprised of α_x chain (CD11c) with β2 chain (CD18). CR4 obtains high affinity to endothelial ligands by extensive conformation changes in both α and β subunits (Alon., 2010). CR4 binding upon ligands thereby, have effect on the immune response such as leukocyte migration and cytokine production. The conformational change can happen because CR4 has a very flexible conformation. And flexible conformation is possible because CR4 has a special contact between the α_x (I) domain and β-propeller domain of α_x-subunit and βI domain of β_2-subunit. Also the α_x I domain of CR4 to recognize ligands and β-propeller domain as having an important role in ligand binding (Xie et al., 2010; Chen et al., 2010). The *CD11c* located in exon15 rs2230429 (Pro517Arg) may play an important role in ligand binding of CR4 through the amino acid of rs2230429 that belongs to β-propeller domain of α_x-subunit as part of FG-GAP (http://www.ncbi.nlm.nih.gov/Structure/cdd/cdd.shtml, cl15299). The rs2230429 C>G (Pro517Arg) of *CD11c* gene have an amino acid sequence change, and thereby, may be affecting the conformation change of CR4. Also, the amino acid of *CD18* located in exon11 rs235326 (Val441VAl) belongs to metal ion dependent adhesion site in von willebrand factor type A domain of β_2-subunit (http://www.ncbi.nlm.nih.gov/Structure/cdd/cdd.shtml, cd00198) and may play an important role in ligand binding.

Also, the frequency of the mutated allele and homozygote genotype of single nucleotide polymorphism (SNP) in exon 15 of *CD11c* gene was significant higher in crohn's disease than in controls **(**Frenzel et al., 2002**)**. It has been reported that the distribution haplotypes of *CD11c* rs2230429 *C>G* and rs2929 *G>A* are associated with gastric ulcer (Hellmig el al., 2005).

The results of this study indicate that the CR4 (CD11c/CD18), *CD11c* gene rs2230429 (Pro517Arg) in exon15, rs2929 in 3'untranslation region (3'UTR) and *CD18* gene rs235326 (Val441VAl) in exon11 polymorphism are positively associated with Behcet's disease (BD). This study found no association between BD and rs2070946 in 5'near gene, rs760456 in intron, and rs684 in 3'UTR of *CD18* gene. The SNPs, rs2230429 and rs2929 of *CD11c* and rs235326 of *CD18* functional significances have not yet been clearly studied. In this study, the homozygous *CD11c* rs2230429 *C/C* and rs2929 *G/G* genotype were significantly higher in BD patients compared to healthy controls (p = 0.0004, OR = 1.8; p = 0.021, OR = 1.5, respectively). The C allele of *CD11c* rs2230429 and G allele of rs2929 were significantly higher in BD patients compared to healthy controls (p <.0001, OR = 1.7; p = 0.013, OR = 1.4, respectively). The significant differences were observed in the distribution of the haplotypes of *CD11c* rs2230429 *C>G* and rs2929 *G>A* SNPs among BD patients and controls. This study combined CD11c rs2230429 *C>G* and rs2929 *G>A* SNPs and obtained results that the frequency of BD patients with *CD11c* rs2230429 *C/C* genotype and rs2929 *G/G* genotype was significantly higher than in healthy controls. The frequency of *CD18* rs235326 *C/C* genotype and C allele were significantly higher in BD than controls

groups (p = 0.002, OR = 1.7; p = 0.001, OR = 1.6, respectively). This study combined the genotype of 4SNPs in CD18 gene, and obtained the result that the frequency of BD patients with *CD18* rs2070946 *A/A* and rs235326 *C/C* genotype was significantly higher than in healthy controls. Although the *CD18* rs235326 (Val441Val) does not have an amino acid change, the SNP can have a different structural variation of mRNA depending on base sequence. And *CD18* rs235326 may possess different biologic functions through the heterogenic mRNA types involved in mRNA synthesis, maturation, transport, translation, or degradation (Shen et al., 1999; Koch et al., 2001). Also, because 3' UTR contribute highly to mRNA safety and gene expression and protein translation rate (Chaterjee et al., 2009), this study combined the genotype of *CD18* rs235326 *T*>C (Val441Val) and rs684 *G>A* (3'UTR). This study obtained the result that the frequency of BD patients with or without *CD18* rs235326 *C/C* genotype and with *CD18* rs684 *G/G* genotype were significantly different compared to healthy controls (p = 0.002, OR = 1.7; p = 0.001, OR = 1.6, respectively) (data not shown).

Because CR4 is heterodimer that is comprised of α_x chain (CD11c) with β2 chain (CD18) and the *CD11c* rs2230429 *C>G* and *CD18* rs235326 *T>C*, which plays an important role in ligand binding, this study combined the genotype of *CD11c* rs2230429 *C>G* and *CD18* rs235326 *T>C*. This study obtained the results that the frequency of BD patients with *CD11c* rs2230429 *C/C* genotype and *CD18* rs235326 *C/C* genotype was significantly higher than in healthy controls. Also, this study combined the genotype of *CD11c* rs2230429 *C>G*, rs2929 *G>A* and *CD18* rs235326 *T>C*, obtained the results that

frequency of BD patients with *CD11c* rs2929 *G/G* and *CD11c* rs2230429 *C/C* and *CD18* rs235326 *C/C* genotype was significantly higher compared to healthy controls ($p = 0.005$, OR = 1.9) and the frequency of BD patients with or without *CD11c* rs2929 genotype and *CD11c* rs2230429 *C/G* or *G/G* and *CD18* rs235326 *C/T*, *T/T* was significantly lower compared to healthy controls ($p = 0.011$, OR = 0.4; $p = 0.0001$, OR = 0.3, respectively). CR4 mediated inflammatory response is enhanced by increased CR4 expression and binding to ligands (Walzog et al., 1999; Vorup-Jensen et al., 2005). CR4 (CD11c/CD18) plays an important role in immune response of several inflammatory disease, experimental autoimmune encephalomyelitis and multiple sclerosis by affecting the T cell activation and cytokine production (Bullard et al., 2006). These results suggested that these SNPs of *CD11c* and *CD18* might be important in BD. Our results provide indirect evidence that suggest SNPs mutation of *CD11c*/*CD18* might change the form and function of CR4.

Also, this study analyzed the relationship between the genotypes of these genes and various clinical parameters such as oral and genital ulcers, ocular lesions, arthritis, vascular involvement, gastrointestinal involvement and neurologic involvement. The SNPs, rs2230429 and rs2929 of *CD11c* and rs235326 of *CD18* had significant associations with the clinical parameters, excluding arthritis and vascular; gastrointestinal involvement; vascular and gastrointestinal involvement, respectively. Interestingly, the BD patients with arthritis were significantly higher than without arthritis in rs2929 *G/G* genotype. The frequency of haplotype rs2230429G - rs2929G was significantly higher in BD patients with arthritis than without arthritis, also.

The CD11c expression increased by high extracellular levels of surviving in rheumatoid arthritis (RA) patients (Mera et al., 2008). Especially, *CD11c* might be association with inflammatory of arthritis through the rs2929 located in 3'UTR of *CD11c* might be contribute to gene expression and mRNA safety of *CD11c.* Although the sample size is small, the frequency of major genotype rs2929 *G/G* was significantly lower in BD patients with neurologic involvement compared to those without neurologic involvement.

There are the frequencies of genotype of *CD11c* and *CD18* gene SNPs in other ethnic groups (Table 8). The genotype frequency of SNPs was different in other ethnic groups. But the number of the population who was investigated in the International HapMap Project is small.

It has been suggested that CR4 (CD11c/CD18) plays a role in susceptibility to BD. No concrete evidence shows whether CR4 (CD11c/CD18) are directly involved in induction or regulation of BD, but CR4 (CD11c/CD18) promote the interaction between endothelium and leukocytes and subsequent migration of the leucocytes to the inflammation site. Genetic variants in exon11 (Pro517Arg) or 3'UTR of *CD11c* and exon11 (Val441Val) of *CD18* may have a role in the risk of developing BD.

In conclusion, this study suggests that the *CD11c* rs2230429*C/C*, rs2929*G/G* and *CD18* rs235326*C/C* genotypes reveal higher susceptibility to BD. Moreover, the haplotype of *CD11c* rs2230429*C* - rs2929*G* and *CD18* rs2070946*A* - rs235326*C* - rs760456*G* - rs684*G* enhances susceptibility to BD. The haplotype of *CD11c* rs2230429*G* - rs2929*A* and *CD18* rs2070946*A* - rs235326*T* - rs760456*C* - rs684*G*

reduces the susceptibility of BD. As mentioned above, these results suggested that the polymorphisms, rs2230429 and rs2929 of *CD11c* and rs235326 of *CD18,* may be helpful in discovering inflammatory development of BD through a contribute gene factor to BD. Further study may be needed to identify the genetic variation of the polymorphisms on a large scale and to measure CR4 expression levels in BD in order to determine whether or not the polymorphisms affect the function or expression of CR4.

Table 1. Clinical characteristics of patients with Behcet's disease

Clinical features	Total (%)	Male (%)	Female (%)
BD patients	305	148 (48.5)	157 (51.5)
Age (years)		19 - 62	16 - 64
Major symptoms			
Oral ulcers	293 (96.1)	144 (97.3)	149 (95.0)
Skin lesions	262 (85.9)	129 (87.2)	133 (84.7)
Genital ulcers	238 (78.0)	107 (72.3)	131 (83.4)
Ocular lesions	186 (61.0)	97 (65.6)	89 (56.7)
Minor symptoms			
Arthritis	95 (30.1)	44 (30.0)	51 (32.5)
Vascular involvement	35 (11.5)	30 (20.1)	5 (3.2)
Gatrointestinal involvement	18 (5.9)	9 (6.1)	9 (5.7)
Neurologic involvement	10 (3.3)	3 (2.0)	7 (4.5)

Table 2. Genotype and allele frequencies of *CD11c* and *CD18* genes in BD

	rs number		BD n=305 (%)	Controls n=266 (%)	*p*	OR (95% CI)	Permutation *p*-value
CD11c							
Exon15 (*1550C>G* Pro517Arg)	rs2230429	*C/C*	193 (63.3)	129 (48.5)	0.0004[a]	1.8 (1.31-2.56)	
		C/G	99 (32.4)	113 (42.5)			
		G/G	13 (4.3)	24 (9.0)	0.021[b]	0.4 (0.22-0.90)	
		C : G	0.80 : 0.20	0.70 : 0.30	<.0001	1.7 (1.31-2.26)	0.001
3'UTR (*80G>A*)	rs2929	*G/G*	166 (54.4)	119 (44.7)	0.021[a]	1.5 (1.06-2.05)	
		G/A	117 (38.4)	118 (44.4)			
		A/A	22 (7.2)	29 (10.9)			
		G : A	0.74 : 0.26	0.67 : 0.33	0.013	1.4 (1.07-1.78)	
CD18							
5'near gene (*-4+7372A>G*)	rs2070946	*A/A*	249 (81.7)	200 (75.2)	0.061[a]	1.5 (0.98-2.19)	
		A/G	51 (16.7)	62 (23.3)			
		G/G	5 (1.6)	4 (1.5)			
		A : G	0.90 : 0.10	0.86 : 0.14			
Exon11 (*1323T>C* Val441Val)	rs235326	*C/C*	203 (66.5)	144 (54.1)	0.002[a]	1.7 (1.20-2.36)	
		C/T	92 (30.2)	104 (39.1)			
		T/T	10 (3.3)	18 (7.8)	0.054[b]	0.5 (0.21-1.03)	
		C : T	0.82 : 0.18	0.74 : 0.26	0.001	1.6 (1.20-2.10)	0.007
Intron (*784C>G*)	rs760456	*C/C*	68 (22.3)	69 (25.9)			
		C/G	150 (49.2)	134 (50.4)			
		G/G	87 (28.5)	63 (23.7)			
		C : G	0.47 : 0.53	0.51 : 0.49			
3'UTR (*122G>A*)	rs684	*G/G*	245 (80.3)	217 (81.6)			
		G/A	56 (18.4)	46 (17.3)			
		A/A	4 (1.3)	3 (1.1)			
		G : A	0.90 : 0.10	0.90 : 0.10			

[a] *p* value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of each major homozygote versus all others combined between the BD patients and control groups.

[b] *p* value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of each minor homozygote versus all others combined between the BD patients and control groups.

Table 3. The genotype distribution *CD11c* rs2230429 and rs2929 of different clinical feature of BD

	n	*CD11c* rs2230429 *C>G*		*p*	OR (95% CI)	*CD11c* rs2929 *G>A*		*p*	OR (95% CI)
		C/C (%)	*C/G* (%)+ *G/G* (%)			*G/G* (%)	*G/A* (%)+ *A/A*(%)		
Controls	266	129 (48.5)	113 (42.5) + 24 (9.0)			119 (44.7)	118 (44.4) + 29 (10.9)		
BD	305	193 (63.3)	99 (32.4) + 13 (4.3)	0.0004	1.8 (1.31-2.56)	166 (54.4)	117 (38.4) + 22 (7.2)	0.021	1.5 (1.06-2.05)
Oral ulcers	293	185 (63.1)	95 (32.4) + 13 (4.5)	0.0005	1.8 (1.30-2.55)	158 (53.9)	115 (39.3) + 20 (6.8)	0.030	1.4 (1.03-2.02)
Skin lesion	262	167 (63.8)	85 (32.4) + 10 (3.8)	0.0004	1.9 (1.32-2.64)	143 (54.6)	102 (38.9) + 17 (6.5)	0.024	1.5 (1.05-2.09)
Genital ulcers	238	155 (65.1)	72 (30.3) + 11 (4.6)	0.0002	2.0 (1.38-2.84)	129 (54.2)	93 (39.1) + 16 (6.7)	0.034	1.5 (1.03-2.08)
Ocular lesions	186	120 (64.5)	55 (29.6) + 11 (5.9)	0.0008	1.9 (1.31-2.84)	101 (54.3)	69 (37.1) + 16 (8.6)	0.045	1.5 (1.01-2.14)
Arthritis	95	54 (56.8)	34 (35.8) + 7 (7.4)			63 (66.3)	25 (26.3) + 7 (7.4)	0.0003	2.4 (1.49-3.97)
Vascular	35	23 (65.7)	12 (34.3) + 0 (0.0)			22 (62.9)	11 (31.4) + 2 (5.7)	0.043	2.1 (1.01-4.32)
Gastrointestinal lesions	18	13 (72.2)	5 (7.8) + 0 (0.0)	0.051	2.8 (0.96-7.96)	11 (61.1)	6 (33.3) + 1 (5.6)		
Neurologic involvement	10	7 (70.0)	3 (30.0) + 0 (0.0)			1 (10.0)	7 (70.0) + 2 (20.0)	0.030	0.1 (0.02-1.10)

p value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of each major homozygote versus all others combined between the BD patients and control groups.

Table 4. The genotype distribution of *CD18* rs235326 of different clinical feature of BD

	n	Genotype frequency		*p*	OR (95% CI)
		C/C (%)	*C/T* (%) + *T/T* (%)		
Controls	266	144 (54.1)	104 (39.1) + 18 (7.8)		
BD	305	203 (66.5)	92 (30.2) + 10 (3.3)	0.002	1.7 (1.20-2.36)
Oral ulcers	293	194 (66.2)	89 (30.4) + 10 (3.4)	0.004	1.6 (1.16-2.34)
Skin lesion	262	173 (66.0)	81 (30.9) + 8 (3.1)	0.005	1.6 (1.16-2.340
Genital ulcers	238	166 (69.8)	65 (27.3) + 7 (2.9)	0.0003	1.9 (1.35-2.82)
Ocular lesions	186	122 (65.6)	57 (30.6) + 7 (3.8)	0.015	1.6 (1.10-2.38)
Arthritis	95	66 (69.5)	26 (27.4) + 3 (3.1)	0.009	1.9 (1.17-3.17)
Vascular	35	24 (68.6)	10 (28.6) + 1 (2.8)		
Gastrointestinal lesions	18	8 (44.4)	9 (50.0) + 1 (5.6)		
Neurologic involvement	10	9 (90.0)	1 (10.0) + 0 (0.0)	0.025	7.6 (0.95-61.03)

p value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of a major homozygote versus all others combined between the BD patients and control groups.

Table 5. The genotype distribution of *CD11c* rs2929 of different clinical feature of BD

	n	*CD11c rs2929*			*p*
		G/G (%)	*G/A* (%)	*A/A*(%)	
BD	305	166 (54.4)	117 (38.4)	22 (7.2)	
Oral ulcers					
With	293	158 (53.9)	115 (39.3)	20 (6.8)	
Without	12	8 (66.6)	2 (16.7)	2 (16.7)	0.181
Skin lesion					
With	262	143 (54.6)	102 (38.9)	17 (6.5)	
Without	43	23 (53.5)	15 (34.9)	5 (11.6)	0.468
Genital ulcers					
With	238	129 (54.2)	93 (39.1)	16 (6.7)	
Without	67	37 (55.2)	24 (35.8)	6 (9.0)	0.773
Ocular lesions					
With	186	101 (54.3)	69 (37.1)	16 (8.6)	
Without	119	65 (54.7)	48 (40.3)	6 (5.0)	0.478
Arthritis					
With	95	63 (66.3)	25 (26.3)	7 (7.4)	
Without	210	103 (49.1)	92 (43.8)	15 (7.1)	0.012
Vascular					
With	35	22 (62.9)	11 (31.4)	2 (5.7)	
Without	270	144 (53.3)	106 (39.3)	20 (7.4)	0.567
gastrointestinal lesions					
With	18	11 (61.1)	6 (33.3)	1 (5.6)	
Without	287	155 (54.0)	111 (38.7)	21 (7.3)	0.837
Neurologic involvement					
With	10	1 (10.0)	7 (70.0)	2 (20.0)	
Without	295	165 (55.9)	110 (37.3)	20 (6.8)	0.012

p value was derived from chi-square statistics from simple 2 X 3 tables based on the frequency of a major homozygote versus each other genotypes between the clinical characteristics with and without.

Table 6. Haplotype frequencies of *CD11c* and *CD18* in BD and Controls

Haplotype	BD	Controls	*p*	OR (95%CI)	Permutation *p*-value
CD11c rs2230429 *C>G* - rs2929*G>A*					
C – G	0.588	0.477	0.0002	1.6 (1.24-1.98)	0.001
C – A	0.207	0.221	0.571	0.9 (0.69-1.22)	
G – G	0.148	0.193	0.044	0.7 (0.53-0.99)	
G – A	0.057	0.110	0.001	0.5 (0.32-0.76)	0.004
CD18 rs2070946 *A>G* - rs235326 *T>C*- rs760456 *C>G* - rs684 *G>A*					
A – C – C – G	0.370	0.354	0.466	1.1 (0.86-1.40)	
A – C – G – G	0.283	0.213	0.007	1.5 (1.11-1.91)	
A – T – G – G	0.119	0.139	0.268	0.8 (0.56-1.17)	
A – C – G – A	0.067	0.055	0.316	1.3 (0.76-2.30)	
A – T – C – G	0.034	0.073	0.003	0.4 (0.26-0.77)	0.012
G – C – G – G	0.037	0.039	0.440	0.8 (0.43-1.44)	
A – C – C – A	0.026	0.034	0.545	0.8 (0.44-1.54)	
G – C – C – G	0.026	0.033	0.355	0.7 (0.39-1.54)	
G – T – G –G	0.022	0.041	0.122	0.5 (0.24-1.19)	

p value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of each haplotype versus all others combined between the BD patients and control groups.

Table 7. Haplotype frequencies of *CD11c* in BD patients with arthritis and without arthritis

Haplotype	Arthritis		*p*	OR (95%CI)	Permutation *p*-value
	With	Without			
CD11c rs2230429 *C>G* - rs2929*G>A*					
C – G	0.581	0.548	0.937	1.0 (0.69-1.39)	
C – A	0.167	0.232	0.065	0.7 (0.42-1.03)	
G – G	0.214	0.125	0.005	1.9 (1.21-3.00)	0.016
G – A	0.039	0.058	0.319	0.6 (0.28-1.51)	

p value was derived from chi-square statistics from simple 2 X 2 tables based on the frequency of a major homozygote versus all others combined between the arthritis with and without in BD patients.

Table 8. The frequencies of genotype of *CD11c* and *CD18* gene in other ethnic groups

SNPs	[1)]Korean	[2)]Japanese	[2)]Chinese	[2)]European	[2)]African	[3)] Germany
CD11c						
rs2230429 *C>G* (Exon15)	n=266					n=162
C/C	129 (48.5)	-	-	-	-	71 (44.0)
C/G	113 (42.5)	-	-	-	-	72 (44.0)
G/G	24 (9.0)	-	-	-	-	19 (12.0)
rs2929 *G>A* (3'UTR)		n=45	n=45	n=59	n=60	n=165
G/G	119 (44.7)	24 (53.3)	32 (71.1)	31 (52.5)	13 (21.7)	106 (63.0)
G/A	118 (44.4)	16 (42.2)	10 (22.2)	22 (37.3)	36 (60.0)	56 (33.0)
A/A	29 (10.9)	2 (4.4)	3 (6.7)	6 (10.2)	11 (18.3)	3 (4.0)
CD18						
rs2070946 *A>G* (5' near gene)		n=44	n=45	n=59	n=59	
A/A	200 (75.2)	37 (84.1)	32 (71.1)	31 (52.5)	44 (74.6)	-
A/G	62 (23.3)	5 (11.4)	13 (28.9)	27 (45.8)	15 (25.4)	-
G/G	4 (1.5)	2 (4.5)	0 (0.0)	1 (1.7)	0 (0.0)	-
rs235326 *T>C* (Exon11)		n=44	n=45	n=59	n=60	
C/C	144 (54.1)	18 (40.9)	25 (55.6)	33 (55.9)	58 (96.7)	-
C/T	104 (39.1)	20 (45.5)	18 (40.0)	22 (37.3)	2 (3.3)	-
T/T	18 (7.8)	6 (13.6)	2 (4.4)	4 (6.8)	0 (0.0)	-
rs760456 *C>G* (Intron)		n=44	n=45	n=59	n=59	
C/C	69 (25.9)	7 (15.9)	9 (20.0)	12 (20.3)	11 (18.6)	-
C/G	134 (50.4)	23 (52.3)	24 (53.3)	22 (37.3)	28 (47.5)	-
G/G	63 (23.7)	14 (31.8)	12 (26.7)	25 (42.4)	20 (33.9)	-
rs684 *G>A* (3'UTR)		n=42	n=45	n=59	n=55	
G/G	217 (81.6)	40 (95.2)	36 (80.0)	38 (64.4)	40 (72.7)	-
G/A	46 (17.3)	2 (4.8)	9 (20.0)	19 (32.2)	13 (23.6)	-
A/A	3 (1.1)	0 (0.0)	0 (0.0)	2 (3.4)	2 (3.6)	-

[1)] In this study [2)] Http://www.hapmap.org/ [3)] Hellmig et al., 2005

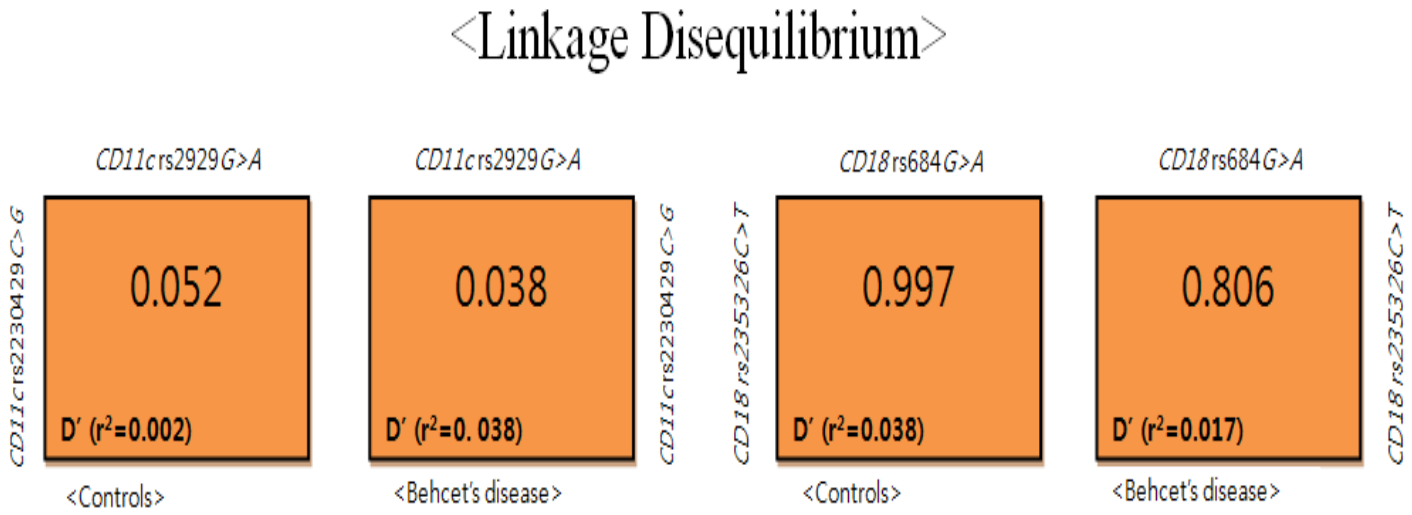

Figure 1. Linkage disequilibrium coefficient (| D' | and r^2) between 2SNPs of *CD11c* and 2SNPs of *CD18*

CD11c	Behcet's disease n=305, %	Controls n=266, %
[1] rs2230429 *C/C* (+) and rs2929 *G/G* (+)	36.0	22.9
[2] rs2230429 *C/C* (-) and rs2929 *G/G* (-)	18.4	29.7
Others	45.6	47.4

p: Behcet's disease vs. Controls

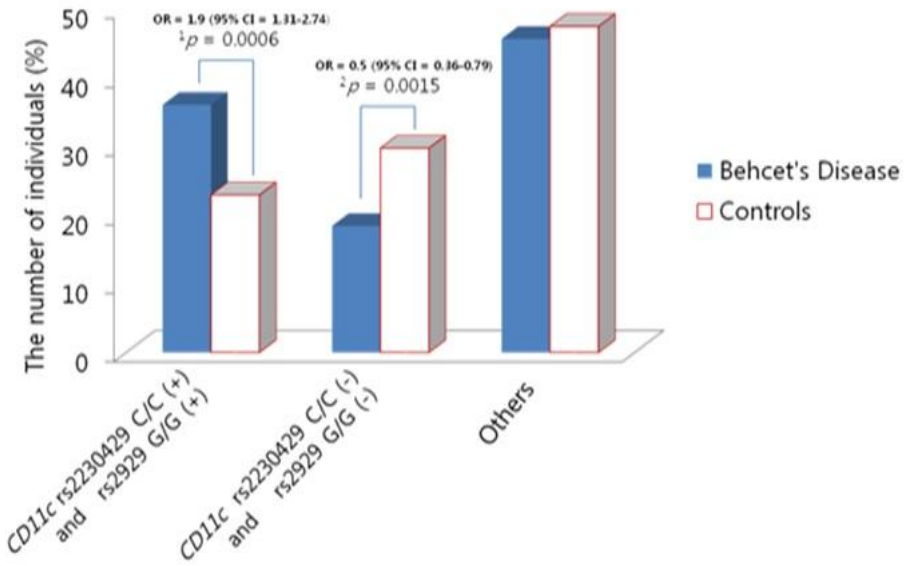

Figure 2. *CD11c* rs2230429*C>G* and rs2929*G>A* combined genotype

· *CD11c* rs2230429 *C/C* (+) : C/C
· *CD11c* rs2230429 *C/C* (-) : C/G, G/G
· *CD11c* rs2929 *G/G* (+) : G/G
· *CD11c* rs2929 *G/G* (-) : G/A, A/A

CD18	Behcet's disease n=305, %	Controls n=266, %
[1] rs2070946 *A/A* (+) and rs235326 *C/C* (+)	56.0	43.2
[2] rs2070946 *A/A* (-) and rs235326 *C/C* (-)	7.9	13.9
Others	36.1	46.9

p: Behcet's disease vs. Controls

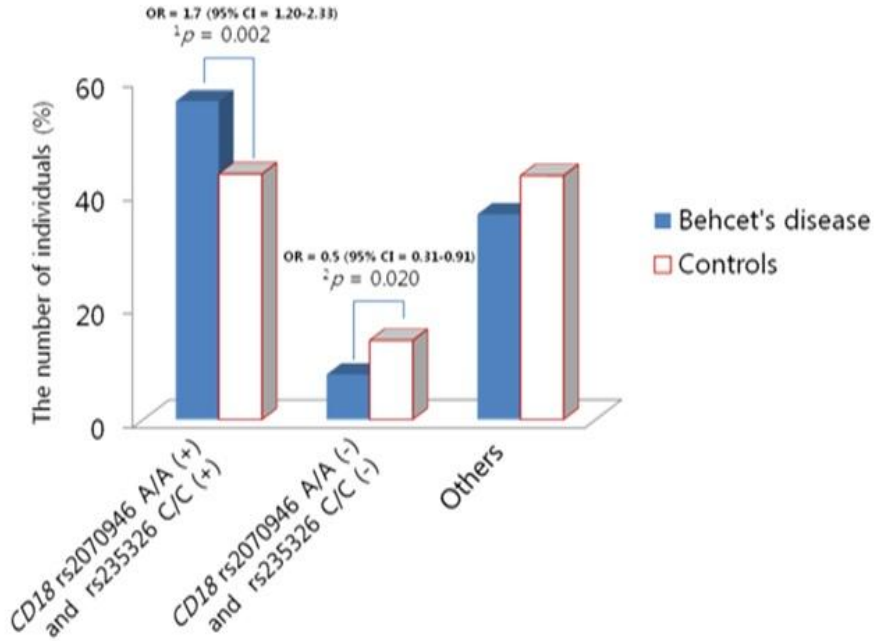

Figure 3. *CD18* rs2070946*A*>*G* and rs235326*T*>*C* combined genotype

· *CD18* rs2070946 *A/A* (+) : A/A
· *CD18* rs2070946 *A/G* (-) : A/G, G/G
· *CD18* rs235326*C/C* (+) : C/C
· *CD18* rs235326 *C/C* (-) : C/T, T/T

	BD n=305, %	Controls n=266, %
[1] *CD11c* rs2230429 *C/C*(+) and *CD18* rs235326*C/C*(+)	40.3	28.6
[2] *CD11c* rs2230429 *C/C*(-) and *CD18* rs235326*C/C*(-)	10.5	25.9
Others	49.2	45.5

p: Behcet's disease vs. Controls

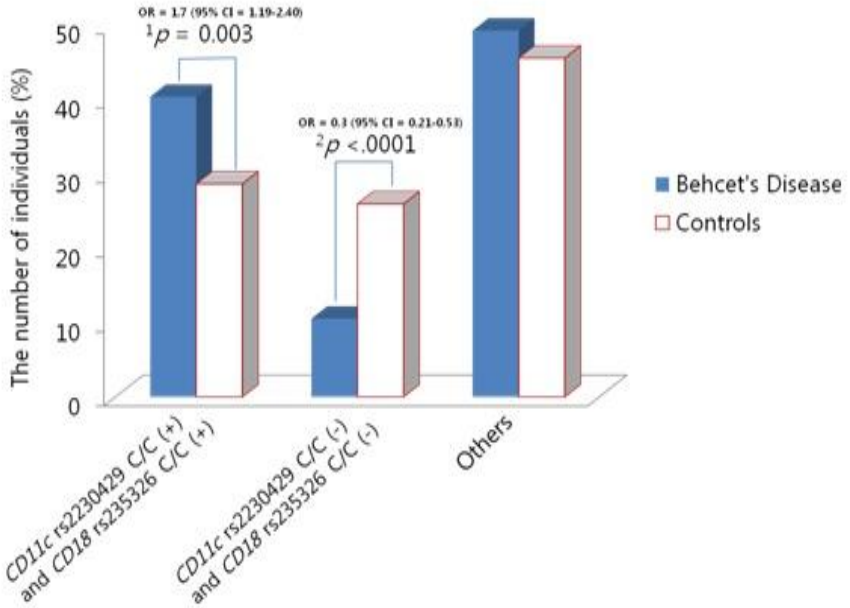

Figure 4. *CD11c* rs2230429*C>G* and *CD18* rs235326*T>C* combined genotype

· *CD11c* rs2230429 *C/C* (+) : C/C
· *CD11c* rs2230429 *C/C* (-) : C/G, G/G
· *CD18* rs235326*C/C* (+) : C/C
· *CD18* rs235326 *C/C* (-) : C/T, T/T

	BD n=305, %	Controls n=266, %
[1] *CD11c* rs2230429 *C/C* (+) and rs2929 *G/G* (+) and *CD18* rs235326 *C/C* (+)	22.6	13.6
[2] *CD11c* rs2230429 *C/C* (-) and rs2929 *G/G* (+) and *CD18* rs235326 *C/C* (-)	4.9	10.5
[3] *CD11c* rs2230429 *C/C* (-) and rs2929 *G/G* (-) and *CD18* rs235326 *C/C* (-)	5.6	15.4
Others	66.9	60.5

p: Behcet's disease vs. Controls

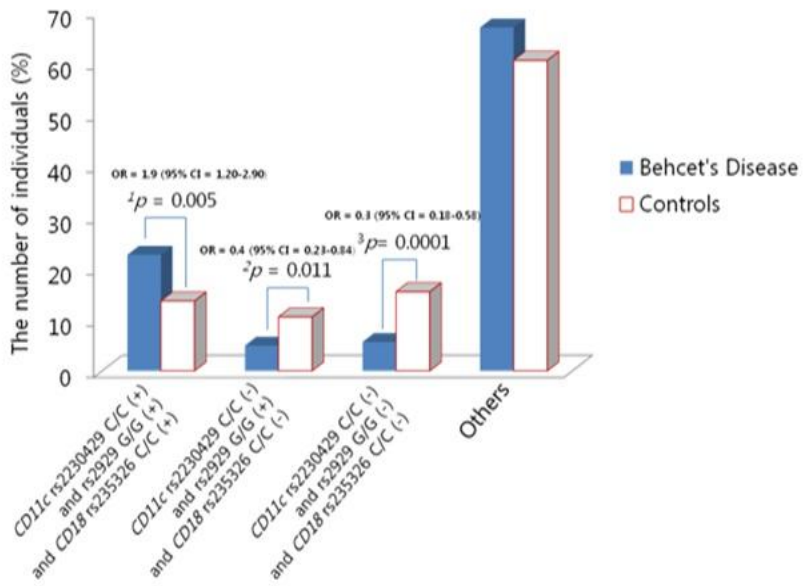

Figure 5. *CD11c* rs2230429*C>G*, rs2929*G>A* and *CD18* rs235326*T>C* combined genotype

· *CD11c* rs2230429 *C/C* (+) : C/C
· *CD11c* rs2230429 *C/C* (-) : C/G, G/G
· *CD11c* rs2929 *G/G* (+) : G/G
· *CD11c* rs2929 *G/G* (-) : G/A, A/A
· *CD18* rs235326*C/C* (+) : C/C
· *CD18* rs235326 *C/C* (-) : C/T, T/T

Web Resources

Accession numbers and URLs for data presented herein are as follows:

International HapMap Project, http://www.hapmap.org/

Conserved Domains Database (CDD),

http://www.ncbi.nlm.nih.gov/Structure/cdd/cdd.shtml

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Abstract

Association between *CR4*(*CD11c/CD18*) Gene Polymorphisms and Behcet's Disease

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Objectives: Behcet's disease (BD) is a chronic and multisystemic inflammatory disease. The pathogenesis cause of BD is still uncertain, but the interaction between leukocyte adhesion and integrins, leukocyte migration into inflammatory site are important responses in the development of inflammatory in BD. Complement receptor type-4 (CR4, CD11c/CD18) is a member of the β2-integrins and plays important role in leukocyte migration into the inflammatory site. This study investigated *CR4* (*CD11c/CD18*) polymorphisms association with BD.

Methods: The single nucleotide polymorphisms (SNPs) in the *CD11c* (rs2230429*C*>*G*, rs2929*G*>*A*) and *CD18* (rs2070946*A*>*G*, rs235326*T*>*C*, rs760456*C*>*G*, rs684*G*>*A*) were genotyped using PCR-RFLP among 305 BD patients and 266 healthy controls in Korean.

Results: The homozygous *CD11c* rs2230429*C/C* genotype and rs2929*G/G* genotype were significantly higher in BD patients than in healthy controls (p = 0.0004, OR = 1.8, 95% CI = 1.31-2.56; p = 0.021 OR = 1.8, 95% CI = 1.06-2.05, respectively). The homozygous *CD18* rs235326 *C/C* genotype was significantly higher in BD patients than in healthy controls (p = 0.002, OR = 1.7, 95% CI = 1.20-2.36). The allele frequencies of *CD11c* rs2230429 *C* (p = <.0001, OR = 1.7, 95% CI = 1.31-2.26), rs2929 *G* (p = 0.013, OR = 1.4, 95% CI = 1.07-1.78) and CD18 rs235326 *C* (p = 0.001, OR = 1.6, 95% CI =

1.20-2.10) were associated with the increased risk of BD. There was no significant difference in the genotype and allele frequency of the *CD18* rs2070946*A>G*, rs760456*C>G* and rs684*G>A*.

Conclusions: These results suggest that the *CD11c* Pro517Arg (rs2230429), 3'UTR (rs2929) and *CD18* Val441Val (rs235326) polymorphisms are associated with BD.

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