Influences of triggering receptor expressed on myeloid cells-1 genetic polymorphisms on genotypes and phenotypes in Korean patients with inflammatory bowel diseases

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The Master's Thesis
submitted to the Department of Medicine,
the Graduate School of Yonsei University
in partial fulfillment of the requirements for the degree
of Master of Medical Science

Eun Suk Jung

June 2010

This certifies that the Master's Thesis of
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June 2010

ACKNOWLEDGEMENTS

This page is exclusively designed to note my gratitude and respect for those who helped me to complete my thesis. I am deeply indebted to my supervisor Prof. Dr. Jae Hee Cheon for his kind help, guidance, support and encouragement throughout my study. Sincere gratitude goes out to my reviewers, Prof. Dr. Tae Il Kim and Prof. Dr. Jong Rak Choi who had the patience and fortitude to read my thesis and provided constructive criticism to help me defend it. Their guidance not only improved my dissertation but also will benefit my future works. I also sincerely thank my colleagues, Kyong Joo Lee, Hyun Jung Lee, Hui Won Jang, Young Eun Chon and Kyu Sik Jung for their supports. Finally, this thesis would not have been possible without my family. This small thesis is devoted to God.

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ABSTRACT

Influences of triggering receptor expressed on myeloid cells-1 genetic polymorphisms on genotypes and phenotypes in Korean patients with inflammatory bowel diseases

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Triggering receptor expressed on myeloid cells-1 (TREM-1) has been shown to play a crucial role in the propagation of inflammatory responses. Recent studies have reported that TREM-1 expression is up-regulated in patients with inflammatory bowel disease (IBD). We hypothesized that *TREM-1* genetic polymorphisms might be associated with IBD development and its phenotypes. We genotyped three *TREM-1* single nucleotide polymorphisms (SNPs, rs2234237, rs3789205, and rs9471535) and evaluated the relationships between these SNPs and IBD genotypes and phenotypes. We found that *TREM-1* SNPs are significantly associated with the development of intestinal Behcet's disease, and in particular with skin involvement and risk of azathioprine use. However, *TREM-1* SNPs were not significantly associated with the development of Crohn's disease or ulcerative colitis. The results of our study suggest that *TREM-1* SNPs may play a significant role in the development of intestinal Behcet's disease and may have modest effects on disease severity.

Key words: Triggering receptor expressed on myeloid cells-1, genetic polymorphism, inflammatory bowel disease, ulcerative colitis, Crohn's disease, intestinal Behcet's disease

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I. INTRODUCTION

Inflammatory bowel disease (IBD) is a chronic intestinal disease with repeated episodes of relapse and remission due to unknown causes.[1-4] The prevalence and incidence of IBD differ among regions. IBD is most common in people of European descent who live in North America and North Europe. Recently, the number of IBD patients has been rapidly increasing in Korea due to the westernization of the Korean diet, improvement of hygiene status, and reduction of infectious disease.[5] In contrast, another form of multisystemic, chronic, relapsing intestinal inflammatory disorder, intestinal Behcet's disease (BD), is most prevalent in Eastern Mediterranean countries and the eastern rim of Asia including Korea.[6]

Although the pathogenesis of IBD, including intestinal BD, is still unclear, genetic and environmental factors are thought to be involved.[7-11] Additional evidence of genetic influences on IBD development include higher frequency in identical twins than fraternal twins, people with a family history of IBD, and differences in prevalence of IBD between races.[12,13] IBD is considered an autoimmune disease caused by abnormal immune reaction to intestinal bacteria that occurs in patients with genetic susceptibilities.[14,15]

Triggering receptor expressed on myeloid cell-1 (TREM-1), a 30-kDA

immunoglobulin superfamily expressed on blood neutrophils and a subset of monocytes, has been shown to play a crucial role in the propagation of local or systemic inflammatory responses.[16] Elevated levels of sTREM-1 have been observed in several infectious diseases.[17,18] A soluble form of TREM-1 (sTREM-1) was observed and identified at significant levels in the sera of patients with sepsis, and sTREM-1 level appeared to be the most helpful parameter in differentiating patients with sepsis from those with systemic inflammatory response syndrome.[19] In addition, an elevated level of sTREM-1 in bronchoavelolar lavage fluid was found to be a strong predictor of pneumonia.[20] As for gastroenterological diseases, pancreatic tissue *TREM-1* mRNA expression was correlated with the severity of pancreatitis,[21] and sTREM-1 level in gastric juice was also correlated with the degree of gastric inflammation scores.[22]

Recently, it was reported that TREM-1 is normally under expressed in the human intestine, which causes immune tolerance to intestinal bacteria and antigens,[23] although TREM-1 is up-regulated in the intestines of patients with IBD.[24] We previously reported that sTREM-1 levels are up-regulated in the sera of IBD patients and correlates with disease activity in patients with ulcerative colitis.[25] However, it remains unknown whether genetic polymorphisms of *TREM-1* influence on genotypes and phenotypes in IBD patients. We hypothesized that genetic polymorphisms of *TREM-1* might be associated with the development of IBD and be correlated with disease phenotypes in IBD. Accordingly, we analyzed *TREM-1* single nucleotide polymorphisms (SNPs) and evaluated the relationships between *TREM-1* polymorphisms and the development and phenotypes of IBD.

II. MATERIALS AND METHODS

1. Study sample

We surveyed 839 unrelated Korean subjects with intestinal diseases, including 202 patients with Crohn's disease (CD), 265 with ulcerative colitis (UC), 138 with intestinal Behcet's disease (BD), and 234 healthy controls. All patients were diagnosed and managed at the gastroenterology clinics of Yonsei University Medical Center, Severance Hospital, Seoul, Republic of Korea. The diagnoses of CD, UC, or intestinal BD were based on established clinical, radiographic, endoscopic, and histopatholgic criteria.[26,27] Exclusion criteria for this study were other autoimmune diseases (systemic lupus erythematosus, rheumatoid arthritis, etc.), chronic hepatitis C, asthma, or coexistence of serious medical or surgical conditions. Healthy controls were recruited among health care center patients who had no gastrointestinal diseases. Demographic and clinical characteristics were obtained by reviewing the medical records of the subjects and through detailed questionnaires.

The CD patients were subdivided into groups according to age at diagnosis, location, and behavior of disease using the Montreal Classifiation.[28] In patients with UC, anatomic location was also subgrouped using the Montreal classification as being ulcerative proctitis (E1), left-sided UC (E2), and extensive UC (E3). Other characteristics including age at diagnosis, gender, extra-intestinal manifestations (EIMs), total follow-up period, the use of immunosuppressive drugs, and the need for surgical intervention were also assessed.

The intestinal BD patients were classified into three groups using colonoscopic findings and extraintestinal systemic manifestations: (i) complete group, (ii) probable group, or (iii) suspected group.[29]

This study was conducted in accordance with the ethical guidelines of the Declaration of Helsinki and was approved by the Institutional Review Board of Yonsei University College of Medicine. Informed written consent was obtained from all subjects.

2. *TREM-1* single nucleotide polymorphism selection and genotyping

We genotyped three single nucleotide polymorphisms (SNPs, rs2234237, rs3789205, and rs9471535). These SNPs were chosen for the study based on a previous Chinese cohort study[30] and Hapmap data (http://www.hapmap.org) regarding the *TREM-1* gene (GenBank access no. DQ217941) in patients and healthy controls. Genomic DNA was isolated from whole blood using commercially available kits (Qiagen, Chatsworth, CA, USA). SNPs were genotyped using the TaqMan® fluorogenic 5′ nuclease assay (Applied Biosystems, Foster City, CA, USA) according to the manufacturer′s protocol. The final volume of each polymerase chain reaction (PCR) was 5 μL, including 20 ng of genomic DNA, 2.5 μL TaqMan® Universal PCR master mix (Applied Biosystems), and 0.22 μL of TaqMan® SNP Genotyping Assay Mix (40x). Amplification conditions were 50°C for 2 min to activate the uracil N-glycosylase and to prevent carryover contamination, then 95°C for 10 min, followed by 40 cycles of 95°C for 15 sec and 60°C for 1 min. The reaction was performed in a 384-well format using a Dual 384-Well GeneAmp® PCR System 9700 (Applied Biosystems). The TaqMan® assay plate was transferred to ABI PRISM 7900 HT Sequence Detection System (Applied Biosystems), where the endpoint fluorescence intensity of each well was read. The calculation of the fluorescence data from each plate was performed using SDS software version 2.3 (Applied Biosystems).

3. Statistical analysis

All SNPs investigated in this study were tested for Hardy-Weinberg equilibrium (HWE) in controls using the χ^2-test. Associations between SNPs and disease susceptibility were determined by comparing allele, genotype, and haplotype frequencies between cases and controls using the χ^2-test or Fisher's exact test. Relationships between *TREM-1* polymorphisms and phenotypes were assessed using logistic regression analysis under a dominant (risk allele homozygotes plus heterozygotes versus wild type allele homozygotes), a recessive (risk allele homozygotes versus heterozygotes plus wild type allele homozygotes), and a codominant (wild type allele homozygotes versus heterozygotes versus risk

allele homozygotes) genetic model. Odds ratios (OR) with 95% confidence intervals (CIs) were determined. To identify significant associations in disease phenotypes and genotypes, we performed comparative analyses within subgroups stratifying UC, CD, or intestinal BD patients by clinical characteristics such as gender, mean age at diagnosis, disease location and behavior, EIMs, history of immunosuppressive drug use, or surgical intervention using logistic regression analysis. *P* values of less than 0.05 were considered statistically significant. The data were analyzed using PASW v.18.0 for Windows (SPSS Inc., Chicago, IL, USA).

III. RESULTS

1. Demographic and clinical characteristics of study sample

Demographic and clinical characteristics of the patients with CD, UC, intestinal BD, and healthy controls who were included in this study are summarized in Table 1.

Table 1. Demographic and clinical characteristics of patients with inflammatory bowel diseases and healthy controls

Characteristics	**CD (n=202)**	**UC (n=265)**	**Intestinal BD (n=138)**	**Control (n=234)**
Mean age at diagnosis (years)[a]	26.0±11.2	36.4±12.4	38.1±11.1	44.0±10.6
≤ 16 years	26 (12.9%)	8 (3.0%)	4 (2.9%)	
17-40 years	156 (77.2%)	167 (63.0%)	83 (60.1%)	
> 40 years	20 (9.9%)	90 (34.0%)	51 (37.0%)	
Gender (male/female)	124 / 78	133 / 132	66 / 72	133 / 101
Follow-up (months) [a]	89.3±51.7	103.6±61.3	89.9±54.7	
Disease localization (%)[b]				
Ileal (L1) ± L4	60 (29.7)			
Colon (L2) ± L4	24 (11.9)			
Ileocolon (L3) ± L4	115 (56.9)			
Proctitis (E1)		64 (24.2)		
Left-sided (E2)		91 (34.3)		
Pancolitis (E3)		110 (41.5)		
Ileocecal region			116(84.1)	
Other colonic region			22 (15.9)	
Disease behavior (%)[b]				
Inflammatory (B1) ± p	81 (40.1)			
Stricturing (B2) ± p	36 (17.8)			
Penetrating (B3) ± p	78 (38.6)			
Clinical subtype of BD (%)				
Complete			107 (77.1)	
Probable			7 (5.1)	
Suspected			24 (17.4)	
Extraintestinal manifestations (%)				
Joint	19 (9.4)	41 (15.5)	48 (34.8)	
Skin	10 (5.0)	25 (9.4)	60 (43.5)	
Eye	11 (5.4)	37 (14.0)	24 (17.4)	
Oral			111 (80.4)	
Genital			42 (30.4)	

Liver		1 (0.4)	
Use of Immunosuppressive drugs (%)	110 (54.5)	69 (26.0)	43 (31.1)
Use of infliximab (%)	36 (17.8)		
History of surgical interventions (%)	80 (39.6)	10 (3.8)	41 (29.7)

CD, Crohn's disease; UC, ulcerative colitis; BD, Behcet's disease.

[a]Mean ± standard deviation

[b]Disease localization and behavior according to Montreal classification (L4, upper gastrointestinal involvement; p, perianal disease)

2. Associations of *TREM-1* genetic polymorphisms with disease susceptibility to inflammatory bowel disease

Three candidate SNPs in *TREM-1* were genotyped to determine their relationships with disease susceptibility to IBD. No significant deviations from HWE were observed in controls. We evaluated the relationships of *TREM-1* genetic polymorphisms with disease susceptibility to CD, UC, and intestinal BD through the analysis of a combined genotype model. We found that *TREM-1* genetic polymorphisms were significantly correlated only with intestinal BD in terms of disease development. Risk allele homozygotes and heterozygotes of rs9471535 at 5′ flanking region of the reference *TREM-1* gene were significantly more frequent in intestinal BD patients compared with healthy controls in dominant model (TT vs. CT/CC, OR=1.606; 95% CI, 1.046 to 2.467, *P*=0.030). Moreover, the risk allele of rs9471535 was significant in codominant model (TT vs. CT vs. CC, *P*=0.026). In rs3789205 at intron 1 and rs2234237 at the exon 1 of *TREM-1* gene, we observed similarly significant results as shown in Table 2.

In CD and UC patients, however, we observed no significant correlations between genotype distributions or combined genotype frequencies of any three SNPs in *TREM-1* (Table 3 and 4).

The linkage disequilibrium (LD) pattern of the three SNPs of *TREM-1* gene is shown in Figure 1. All of the three SNPs were located in the same haplotype block, and were in tight LD (D'>0.99, r^2>0.96).

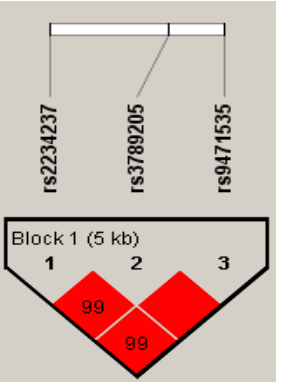

Figure 1. Linkage disequilibrium structure of the three *TREM-1* SNPs in Koreans.

The linkage disequilibrium (LD) patterns of the three SNPs in *TREM-1* gene with D' values are shown.

Table 2. Associations of *TREM-1* SNP genotypes between patients with intestinal Behcet's disease and healthy controls

rs number		genotype	Control (n=234)	BD (n=135)	*P*-value	OR (95% CI)
rs9471535	Dominant	TT	121	54	0.030	1.606 (1.046-2.467)
		CT/CC	113	81		
	Recessive	TT/CT	212	126	0.362	0.688 (0.307-1.541)
		CC	22	9		
	Codominant	TT	121	54	0.026	
		CT	91	72		
		CC	22	9		
rs3789205	Dominant	AA	120	54	0.023	1.637 (1.068-2.510)
		AG/GG	114	84		
	Recessive	AA/AG	212	128	0.474	0.753 (0.345-1.641)
		GG	22	10		
	Codominant	AA	120	54	0.027	
		AG	92	74		
		GG	22	10		
rs2234237	Dominant	TT	116	52	0.020	1.659 (1.080-2.547)
		AT/AA	117	87		
	Recessive	TT/AT	211	128	0.616	0.824 (0.387-1.756)
		TT	22	11		
	Codominat	TT	116	52	0.033	
		AT	95	76		
		AA	22	11		

BD, Behcet's disease; OR, odds ratio; CI, confidence interval.

Table 3. Associations of *TREM-1* SNP genotypes between patients with Crohn's disease and healthy controls

rs number		genotype	Control (n=234)	CD (n=194)	*P*-value	OR (95% CI)
rs9471535	Dominant	TT	121	94	0.502	1.139 (0.778-1.667)
		CT/CC	113	100		
	Recessive	TT/CT	212	181	0.310	0.692 (0.339-1.413)
		CC	22	13		
	Codominant	TT	121	94	0.354	
		CT	91	87		
		CC	22	13		
rs3789205	Dominant	AA	120	93	0.474	1.143 (0.781-1.673)
		AG/GG	114	101		
	Recessive	AA/AG	212	182	0.221	0.635 (0.306-1.319)
		GG	22	12		
	Codominant	AA	120	93	0.259	
		AG	92	89		
		GG	22	112		
rs2234237	Dominant	TT	116	93	0.666	1.087 (0.743-1.591)
		AT/AA	117	102		
	Recessive	TT/AT	211	183	0.210	0.629 (0.303-1.306)
		TT	22	12		
	Codominant	TT	116	93	0.324	
		AT	95	90		
		AA	22	12		

CD, Crohn's disease; OR, odds ratio; CI, confidence interval.

Table 4. Associations of *TREM-1* SNP genotypes between patients with ulcerative colitis and healthy controls

rs number		genotype	Control (n=234)	UC (n=257)	*P*-value	OR (95% CI)
rs9471535	Dominant	TT	121	138	0.660	0.923 (0.648-1.317)
		CT/CC	113	119		
	Recessive	TT/CT	212	232	0.902	1.038 (0.568-1.897)
		CC	22	25		
	Codominant	TT	121	138	0.870	
		CT	91	94		
		CC	22	25		
rs3789205	Dominant	AA	120	138	0.757	0.946 (0.665-1.346)
		AG/GG	114	124		
	Recessive	AA/AG	212	235	0.736	1.107 (0.612-2.003)
		GG	22	27		
	Codominant	AA	120	138	0.853	
		AG	92	97		
		GG	22	27		
rs2234237	Dominant	TT	116	135	0.667	0.925 (0.650-1.318)
		AT/AA	117	126		
	Recessive	TT/AT	211	234	0.738	1.107 (0.612-2.002)
		TT	22	27		
	Codominant	TT	116	135	0.800	
		AT	95	99		
		AA	22	27		

UC, ulcerative colits; OR, odds ratio; CI, confidence interval.

3. Associations of *TREM-1* genetic polymorphisms with intestinal BD phenotypes

We also evaluated the relationships between the *TREM-1* SNPs and phenotypes of intestinal BD (Table 5).

Table 5. Associations of *TREM-1* SNP genotypes with intestinal Behcet's disease phenotypes (univariate analysis)

Disease phenotype	rs number		genotype	Patients without phenotype (%)	Patients with phenotype (%)	*P*-value	OR (95% CI)
Skin	rs9471535	Dominant	TT	39 (50.6)	15 (25.9)	0.004	2.942 (1.406-6.156)
			CT/CC	38 (49.4)	43 (74.1)		
		Recessive	TT/CT	75 (97.4)	51 (87.9)	0.029	5.147 (1.028-25.781)
			CC	2 (2.6)	7 (12.1)		
	rs3789205	Dominant	AA	39 (48.8)	15 (25.9)	0.007	2.727 (1.310-5.677)
			AG/GG	41 (51.3)	43 (74.1)		
		Recessive	AA/AG	77 (96.3)	51 (87.9)	0.095	3.523 (0.870-14.258)
			GG	3 (3.8)	7 (12.1)		
	rs2234237	Dominant	TT	37 (46.3)	15 (25.4)	0.012	2.524 (1.213-5.251)
			AT/AA	43 (53.8)	44 (74.6)		
		Recessive	TT/AT	76 (95.0)	52 (88.1)	0.203	2.558 (0.712-9.182)
			TT	4 (5.0)	7 (11.9)		
Azathioprine use	rs9471535	Dominant	TT	43 (46.2)	11 (26.2)	0.028	2.424 (1.089-5.392)
			CT/CC	50 (53.8)	31 (73.8)		
		Recessive	TT/CT	86 (92.5)	40 (95.2)	0.720	0.614 (0.122-3.090)
			CC	7 (7.5)	2 (4.8)		
	rs3789205	Dominant	AA	43 (44.8)	11 (26.2)	0.039	2.286 (1.031-5.073)
			AG/GG	53 (55.2)	31 (73.8)		
		Recessive	AA/AG	88 (91,7)	40 (95.2)	0.723	0.550 (0.112-2.708)
			GG	8 (8.3)	2 (4.8)		
	rs2234237	Dominant	TT	42 (43.3)	10 (23.8)	0.029	2.444 (1.081-5.525)
			AT/AA	55 (56.7)	32 (76.2)		
		Recessive	TT/AT	88 (90.7)	40 (95.2)	0.504	0.489 (0.101-2.367)

			TT	9 (9.3)	2 (4.8)		
Ileal involvement	rs3789205	Dominant	AA	29 (42.0)	25 (36.2)	0.485	1.276 (0.643-2.532)
			AG/GG	40 (58.0)	44 (63.8)		
		Recessive	AA/AG	67 (97.1)	61 (88.4)	0.049	4.393 (0.898-21.498)
			GG	2 (2.9)	8 (11.6)		

BD, Behcet's disease; OR, odds ratio; CI, confidence interval.

We included all other co-variates (gender, age at diagnosis, and use of immunosuppressive drugs) in a multiple logistic regression analysis in order to adjust for the different clinical manifestations (Table 6).

Table 6. Associations of *TREM-1* SNP genotypes with intestinal Behcet's disease phenotypes (multiple logistic regression analysis)

Disease phenotype	rs number		genotype	*P*-value*	OR (95% CI)
Skin	rs9471535	Dominant	TT	0.004	0.326 (0.152-0.700)
			CT/CC		
		Recessive	TT/CT	0.047	0.192 (0.038-0.975)
			CC		
	rs3789205	Dominant	AA	0.008	0.361 (0.170-0.768)
			AG/GG		
		Recessive	AA/AG	0.064	0.251 (0.058-1.083)
			GG		
	rs2234237	Dominant	TT	0.015	0.392 (0.184-0.832)
			AT/AA		
		Recessive	TT/AT	0.154	0.391 (0.107-1.424)
			TT		
Azathioprine use	rs9471535	Dominant	TT	0.023	0.365 (0.154-0.872)
			CT/CC		
		Recessive	TT/CT	0.556	1.656 (0.309-8.872)
			CC		
	rs3789205	Dominant	AA	0.047	0.417 (0.170-0.987)
			AG/GG		
		Recessive	AA/AG	0.185	3.186 (0.574-17.676)
			GG		
	rs2234237	Dominant	TT	0.034	0.391 (0.164-0.933)
			AT/AA		
		Recessive	TT/AT	0.296	2.374 (0.469-12.023)
			TT		
Ileal involvement	rs3789205	Dominant	AA	0.777	0.900 (0.436-1.860)
			AG/GG		
		Recessive	AA/AG	0.047	0.188 (0.036-0.977)
			GG		

BD, Behcet's disease; OR, odds ratio; CI, confidence interval.

* After adjustment for all other covariates (gender, age at diagnosis, and use of immunosuppressive drugs)

We found that dominant models of all of three SNPs were associated with skin involvement and a higher risk of azathioprine use in intestinal BD. Moreover, recessive model of rs3789205 was associated with ileal involvement of intestinal BD (AA/AG vs. GG, OR=0.188; 95% CI, 0.036 to 0.977, *P*=0.047 in a multiple logistic regression analysis).

IV. DISCUSSION

In this study, we examined the relationships between *TREM-1* genetic polymorphisms and susceptibility to IBD and disease phenotypes. It was previously reported that TREM-1 is underrepresented in the human intestine which results in immune tolerance to intestinal bacteria and antigens,[23] while TREM-1 is upregulated in the intestine of patients with IBD.[24] In an experimental mouse model with colitis, TREM-1-expressing intestinal macrophages contributed to amplify chronic inflammation.[24] Our group also reported that sTREM-1 levels are upregulated in the sera of IBD patients and correlates with disease activity in patients with UC.[25] Furthermore, a study investigating an association between *TREM-1* gene polymorphisms and severe sepsis was conducted in a Chinese cohort.[30] Based on these observations, development and phenotypes of IBD could be different among *TREM-1* SNPs. Therefore, we hypothesized that *TREM-1* genetic polymorphisms might be associated with development of CD and UC. To the authors' knowledge, this is the first attempt to investigate the correlations of *TREM-1* SNPs with IBD genotypes and phenotypes. However, *TREM-1* genetic polymorphisms did not show a significant association with the development of CD and UC. These findings suggest that *TREM-1* genetic polymorphisms are not associated with CD or UC. However, further studies are needed to exclude the possibility that this finding may have been due to the small sample size of this study, or to the exclusion of other unknown SNPs from consideration.

We also examined the relationships between *TREM-1* genetic polymorphisms and susceptibility to intestinal BD. Three common markers showed similarly significant association. The SNP (rs9471535) located within the 5′ flanking region of the reference *TREM-1* gene may impact on the transcriptional activity, and other nonsynonymous variation Ser25Thr (rs2234237) in the second exon of *TREM-1* gene may influence the biologic function of TREM-1. All of the three SNPs were located in the same haplotype block and were in tight LD, which suggests *TREM-1* gene may strongly involved in BD.

Intestinal BD is similar to CD in terms of its clinical features and

therapeutic implications.[31] Both diseases develop at a young age, have similar gastrointestinal and extraintestinal manifestations, and have chronic relapsing and remission courses. Because of these clinical similarities, it is occasionally difficult to distinguish between intestinal BD and CD. Despite similar manifestations in clinical practice, intestinal BD is generally agreed to be a distinct entity.[32,33] Previous genetic studies on BD have shown that *HLA-B*51* carriers were more likely to develop BD, and that the frequency of *HLA-B*51* was much higher in male patients.[34-36] Moreover, *tumor necrosis factor-α-1031* T/C polymorphisms were reported be associated with BD susceptibility.[37] The distribution of *IL-8* gene haplotype was also associated with BD.[38] Recent studies showed that SNPs of *TLR-4*[39] and *IL-17F* may influence the susceptibility of BD.[40] Baranathan et al. reported that *protein tyrosine phosphatase type 22* (PTPN22 620W) was inversely associated with BD susceptibility.[41] Gunesacar et al. reported SNP of the *CD 28* gene was increased in BD patients, whereas the *CTLA-4* gene was decreased.[42] With regard to intestinal BD, however, there have been no reports on genetic polymorphisms. Therefore, a genetic polymorphism study is necessary with respect to intestinal BD. Accordingly, we hypothesized that *TREM-1* genetic polymorphisms might be associated with intestinal BD susceptibility.

To the best of our knowledge, this is the first trial to explore the relationships between genetic polymorphisms and intestinal BD. Our data indicate that all three *TREM-1* SNPs were significantly associated with susceptibility to intestinal BD. According to these results, intestinal BD may differ genetically from other IBDs such as CD or UC. Some groups have claimed that intestinal BD and CD are the same disease with different spectra.[43,44] However, based on our observations, these two diseases should be considered different disease entities. One of the main histological findings of intestinal BD is neutrophil infiltrations around the ulcers. Hayasaki et al. showed that neutrophilic phlebitis may be involved in the pathogenesis of intestinal BD.[45] Because TREM-1 is mainly expressed in neutrophils and monocytes, TREM-1 might contribute to the pathogenesis of intestinal BD. Moreover, we found that *TREM-1* SNPs are associated with skin involvement as well as use of azathioprine as a treatment for

intestinal BD. The association of *TREM-1* SNPs and use of azathioprine suggests that the presence of *TREM-1* SNPs are associated with higher disease activity of intestinal BD. *TREM-1* SNPs, however, are not associated with increased necessity of operation. Moreover, we found that these three SNPs are not correlated with sudden serious complications such as perforation. Therefore, we hypothesize that *TREM-1* SNPs have a modest effect on intestinal BD manifestations. However, there were no significant associations between *TREM-1* SNPs and oral, genital tract, joint, or eye involvement. Further study is warranted to determine the relationships of *TREM-1* SNPs with not only intestinal BD but also systemic BD.

Our study has several limitations. First of all, our study is limited by its small sample size. Because of this small sample size, we were unable to demonstrate strong associations between *TREM-1* genetic polymorphisms and disease phenotypes in recessive disease models. Second, our study was not based on unbiased genome wide association scanning. Finally, our results were obtained in a Korean sample and should be validated in other ethnic groups.

V. CONCLUSION

TREM-1 SNPs have a significant association with the development of intestinal BD, in particular, its skin involvement and use of azathioprine for treatment. However, *TREM-1* SNPs do not have a significant influence on development of CD and UC. The results of our study suggest that *TREM-1* SNPs could play a significant role in the development of intestinal BD and may have modest effects on its disease severity.

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ABSTRACT (IN KOREAN)

한국인 염증성 장질환 환자에서 triggering receptor expressed on myeloid cells-1의 유전자 다형성이 질환 감수성과 표현형에 미치는 영향 분석

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Triggering receptor expressed on myeloid cells-1 (TREM-1)은 중성구나 단핵구에서 발현되는 면역글로불린 종의 하나이다. TREM-1은 면역반응의 진행에 중요한 역할을 하는 것으로 알려져 있다. 최근의 연구에서는 TREM-1이 염증성 장질환과의 연관성도 보고되고 있고 유전적 다형성도 보고되고 있다. 이번 연구에서는 *TREM-1*의 유전적 다형성이 염증성 장질환의 발생 및 표현형과 연관이 있을 것이라는 가설을 세웠다. *TREM-1*의 3가지 단일염기다형성 (rs2234237, rs3789205, and rs9471535)을 유전자 분석하고 3가지 염증성 장질환인 크론병, 궤양성 대장염, 베체트 장염의 감수성과 표현형의 연관성에 대해 살펴보았다. *TREM-1* 단일염기다형성은 크론병과 궤양성대장염과는 의미있는 연관성을 보이지 않았으나, 베체트장염과는 의미있는 연관성을 보였다 (P<0.05). 또한, *TREM-1* 단일염기다형성은 베체트장염의 피부 침범과 azathioprine 사용과도 관련이 있었다. 이 연구 결과들을 통해 *TREM-1* 단일염기다형성이 베체트장염의 유전형과 표현형 발현에 영향을 미침을 시사한다.

핵심되는 말 : Triggering receptor expressed on myeloid cells-1, 유전자 다형성, 염증성 장질환, 궤양성 대장염, 크론병, 베체트 장염