

# The Correlation of Serum IL-12B Expression With Disease Activity in Patients With Inflammatory Bowel Disease

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**Abstract:** Genetic variants in *IL12B*, encoding the p40 subunit common in interleukin-12 (IL-12) and interleukin-23, were identified as the susceptibility loci for inflammatory bowel disease (IBD). This study aimed to identify the correlation of serum IL-12B expression with disease activity in patients with IBD and evaluate the possibility of IL-12B as a biomarker for assessing inflammatory status in IBD.

A total of 102 patients with IBD, including 38, 32, and 32 patients with Crohn's disease (CD), ulcerative colitis (UC), and intestinal Behçet's disease (intestinal BD), respectively, were included. The clinical and laboratory data from the patients were collected at the time of serum IL-12B measurement. Serum IL-12B levels were measured using an enzyme-linked immunosorbent assay.

The median IL-12B levels in patients with CD, UC, and intestinal BD were significantly higher than those in controls (1.87, 2.74, and 2.73 pg/mL, respectively, vs. 1.42 pg/mL, all  $P < 0.05$ ). IL-12B concentrations were associated with disease activity in patients with UC and intestinal BD but not in those with CD. IL-12B levels were increased with increasing disease activity in patients with UC ( $P < 0.001$ ). Likewise, patients with active intestinal BD had higher IL-12B levels than those without active disease ( $P = 0.008$ ). IL-12B levels were correlated with the endoscopic disease activity of UC ( $P = 0.002$ ) and intestinal BD ( $P = 0.001$ ) but not that of CD.

Editor: Shusuke Onishi.

Received: January 23, 2016; revised: April 23, 2016; accepted: April 29, 2016.

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This study was supported by the Korean Health Technology R&D Project through the Korea Health Industry Development Institute (KHIDI), funded by the Ministry of Health and Welfare, Republic of Korea (Grant Number A120176, H113C1345, H112C0130), by the Basic Science Research Program through the National Research Foundation of Korea funded by the Ministry of Science, ICT & Future Planning (NRF-2014R1A1A1008096), and by the "Kiturami" Faculty Research Assistance Program of Yonsei University College of Medicine for 2012 (6-2012-0135).

The authors have no conflicts of interest to disclose.

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ISSN: 0025-7974

DOI: 10.1097/MD.00000000000003772

Serum IL-12B levels were significantly correlated with clinical and endoscopic disease activity in patients with UC and intestinal BD, suggesting its potential use as a biomarker for assessing disease activity in these patients.

(*Medicine* 95(23):e3772)

**Abbreviations:** BD = Behçet's disease, CD = Crohn's disease, CDAI = Crohn's disease activity index, CRP = C-reactive protein, ESR = erythrocyte sedimentation rate, IBD = inflammatory bowel disease, SES-CD = simplified endoscopic activity score for Crohn's disease, UC = Ulcerative colitis.

## INTRODUCTION

Inflammatory bowel disease (IBD) is characterized by chronic idiopathic recurrent inflammation of the gastrointestinal tract.<sup>1</sup> Crohn's disease (CD) and ulcerative colitis (UC) are clinically the two main subtypes of IBD. Intestinal Behçet's disease (BD) caused by the digestive tract involvement of BD has similar clinical and therapeutic characteristics as IBD, and it is also considered an additional subtype of IBD. The pathogenesis of IBD remains unclear. Various environmental and host factors including genetic, epithelial, and immunological factors are involved.<sup>2</sup> Some studies reported that a dysregulated immune response triggered by environmental factors and microbiota results in recurrent inflammation of the gastrointestinal tract in genetically susceptible patients.<sup>3</sup> Subsequently, many studies have focused on finding susceptible variants and their effects on IBD.

Among candidates, genome-wide association studies identified *IL23R* and additional genes involved in Th17 differentiation such as *IL12B*, *JAK2*, *TYK2*, *STAT3*, *CCR6*, *IL2/IL21*, and *TNFSF15* as being associated with susceptibility to IBD.<sup>4–7</sup> Interleukin (IL)-12 and IL-23 are known as proinflammatory cytokines playing significant roles in bridging the innate and adaptive immune systems in patients with IBD.<sup>8,9</sup> IL-12 promotes the differentiation of naive CD4+T cells into Th1 effector cells and is a potent stimulus of natural killer and CD8+T cells.<sup>6</sup>

Biomarkers in IBD are used for making an accurate diagnosis, determining disease activity, and stratifying the risk of an unfavorable course. However, current biomarkers are not disease-specific, but they instead reflect generalized inflammation.<sup>10</sup> Thus, serum markers of acute phase responses such as C-reactive protein (CRP) or the erythrocyte sedimentation rate (ESR) have limitations because of their lack of disease specificity or lower sensitivity. Additionally, although endoscopic evaluation is available, endoscopy is also limited because of its invasiveness and patient discomfort. Thus, serologic biomarkers to appropriately represent disease activity in IBD are warranted.

The relationships between disease activity and serum IL-12B levels in patients with IBD remain unclear. Therefore, we investigated the usefulness of serum IL-12B measurements in assessing disease activity of IBD through evaluating the circulating levels of IL-12B in patients who were diagnosed with CD, UC, or intestinal BD.

## METHODS

### Study Subjects

Between January 2006 and February 2013, a total of 102 patients with IBD including patients with CD, UC, and intestinal BD who visited Severance Hospital in Korea were included. According to the diagnostic criteria for IBD, the patients were diagnosed with CD, UC, or intestinal BD using clinical, radiological, endoscopic, and histological criteria.<sup>11,12</sup> We retrospectively analyzed their clinical data at the time of the initial IBD diagnosis, and their blood samples were collected on the same day. Healthy individuals matched by age and sex with patients with IBD were included in the control group. The study protocol was performed in accordance with the ethical guidelines of the 1975 Declaration of Helsinki. Written, informed consent was obtained from each patient. This study was approved by the Institutional Review Board of Severance Hospital, Yonsei University College of Medicine (4-2014-1012).

### Determination of Disease Activity

Clinical activity was scored using the CD activity index (CDAI) for CD and the guidelines of the American College of Gastroenterology for UC, with higher scores indicating more severe disease.<sup>13,14</sup> A CDAI score of less than 150 indicated quiescent disease, whereas scores of 150 to 250, 250 to 350, >350 denoted mild disease, moderate disease, and severe disease, respectively. The disease activity of intestinal BD was assessed on the basis of the CDAI. Endoscopic disease activity was determined using the Simplified Endoscopic Activity Score for Crohn's disease for CD and the Mayo score for UC and intestinal BD.<sup>15</sup> For the present study, the Simplified Endoscopic Activity Scores for Crohn's disease activity levels were defined as follows: 0 to 3, remission (inactive); 4 to 10, mild activity; 11 to 19, moderate activity; and  $\geq 20$ , high activity.<sup>16</sup>

### Sampling and Measurement of Serum IL-12B

Peripheral blood was obtained from patients with IBD at the time of the initial diagnosis. A single blood sample was taken from healthy controls. Blood was centrifuged at  $1000 \times g$  for 15 min, and serum was isolated and stored at  $-80^{\circ}\text{C}$  until analysis. Levels of serum IL-12B were measured in triplicate using an enzyme-linked immunosorbent assay (Human IL-12 Immunoassay Quantikine; R&D Systems, Minneapolis, MN) following the manufacturer instructions. Briefly, samples were incubated in wells precoated with an IL-12B-specific antibody at room temperature for 2 h. After 3 washes, a peroxidase-conjugated anti-IL-12B antibody was added to the wells, and samples were then incubated for 2 h at room temperature. After 3 washes, substrates were added. After 20-min incubation at room temperature, stop solution was added to terminate the enzyme reaction. Optical density measurements were performed at 450 nm using a microplate reader (Molecular Devices Corporation, Sunnyvale, CA), with the wavelength corrected by comparing and measuring the

absorbance at 540 nm. Serum ESRs and CRP levels were measured via capillary photometry (Test 1 BCL; Alifax, Polverara, Italy) and an immunoturbidimetric assay (Hitachi 7600 P module, Japan), respectively. IL-12B and CRP levels and the ESR were measured at the time of colonoscopic examination.

### Statistical Analysis

Frequencies and percentages were determined for categorical variables. The Kruskal–Wallis test as a nonparametric test or the Mann–Whitney *U* test was used to assess differences between patients with IBD and controls. To analyze the trend of IL-12B levels in accordance with disease activity, the Jonckheere trend test was used. All statistical analyses were performed using SAS (version 9.2, SAS Inc, Cary, NC).  $P < 0.05$  was considered statistically significant.

## RESULTS

### Baseline Characteristics of Patients

A total of 102 patients with IBD with a median age of 30.8 (range, 15.0–70.0) years were studied, and 60 (58.8%) of the patients were men. The median age of the healthy controls ( $n = 12$ ) was 30.2 (range, 27.0–35.0) years, and 8 (66.7%) of the controls were men. CD, UC, and intestinal BD were diagnosed in 38 (37.2%), 32 (31.4%), and 32 (31.4%) patients, respectively. All clinical features of the study subjects are shown in Table 1.

### Serum IL-12B Levels in Patients and Control Subjects

The median concentration of IL-12B by disease regardless of disease activity was as follows: 1.87 (range, 0.08–7.75) pg/mL in CD, 2.74 (range, 0.01–12.0) pg/mL in UC, and 2.73 (range, 0.79–13.0) pg/mL in intestinal BD (Table 1). Compared with the control group, IL-12B levels were statistically different in all 3 disease groups (control vs. CD,  $P = 0.011$ ; control vs. UC,  $P = 0.002$ ; control vs. BD,  $P = 0.009$ ).

### Levels of IL-12B and IBD Disease Activity

Next, differences of IL-12B levels by disease activity were analyzed (Table 2). In patients with CD, IL-12B levels were not correlated with disease activity (Figure 1A). Among patients with CD, IL-12B levels were 1.24 (range, 0.10–1.98) pg/mL in those in remission, 2.86 (range, 0.80–7.75) pg/mL in those with mild disease, 1.09 (range, 0.08–4.63) pg/mL in those with moderate disease, and 3.06 (range, 2.65–4.43) pg/mL in those with severe CD ( $P = 0.280$ ). As a result of subanalysis in accordance with CD behavior, there was no difference in IL-12B levels related to CD behavior according to Montreal classification.

The same comparisons were performed for patients with UC and intestinal BD. A significant correlation was noted between IL-12B levels and disease activity in patients with UC and intestinal BD (Figure 1B and C). The levels of IL-12B in patients with UC were as follows: 1.10 (range, 0.01–10.0) pg/mL among those in remission, 1.18 (range, 0.12–2.77) pg/mL among those with mild disease, 3.19 (range, 2.83–6.56) pg/mL among those with moderate disease, and 4.69 (range, 3.18–12.0) pg/mL among those with severe UC ( $P < 0.001$ ). Among patients with intestinal BD, serum IL-12B levels were 1.11 (range, 0.88–4.24) pg/mL among those in remission, 2.49 (range, 0.79–9.0) pg/mL among those with

TABLE 1. Baseline Characteristics of the Study Population

Variables	Control (n = 12)	CD (n = 38)	UC (n = 32)	BD (n = 32)
Age, y	30.2 (27–35)	27.4 (15–70)	37.0 (16–69)	38.4 (16–63)
Male sex	8 (66.7)	28 (73.7)	20 (62.5)	12 (37.5)
ESR, mm/h	4.5 (0.3–8.4)	67.0 (2.0–120.0)	19.0 (2.0–77.0)	32.3 (0.4–109.0)
CRP, mg/dL	3.7 (2.5–5.0)	14.9 (1.0–143.2)	6.6 (0.4–33.3)	3.9 (0.1–92.7)
IL-12B, pg/mL	1.42 (0.09–4.23)	1.87 (0.08–7.75)	2.74 (0.01–12.0)	2.73 (0.79–13.0)
Montreal classification of CD				
L1 (isolated ileal disease)		9 (23.7)		
L2 (isolated colonic disease)		8 (21.0)		
L3 (ileocolonic disease)		21 (55.3)		
L4 (concomitant upper gastrointestinal tract disease)		3 (7.9)		
P (concomitant perianal disease)		12 (31.6)		
B1 (nonstricturing, nonpenetrating)		26 (68.4)		
B2 (stricturing)		7 (18.4)		
B3 (penetrating)		5 (13.2)		
Montreal classification of extent of UC				
E1 (ulcerative proctitis)			11 (34.4)	
E2 (left-sided UC)			12 (37.5)	
E3 (extensive UC)			9 (28.1)	
Ulcer type of intestinal BD				
Typical				27 (84.4)
Atypical				5 (15.6)
Ulcer location of intestinal BD				
Terminal ileum				19 (59.4)
Ileocecal valve				8 (25.0)
Cecum				3 (9.4)
Ascending colon				2 (6.3)

Variables are expressed as the median (range) or n (%) as appropriate.  
BD = Behçet's disease, CD = Crohn's disease, CRP = C-reactive protein, ESR = erythrocyte sedimentation rate, IL-12B = interleukin-12B, UC = ulcerative colitis.

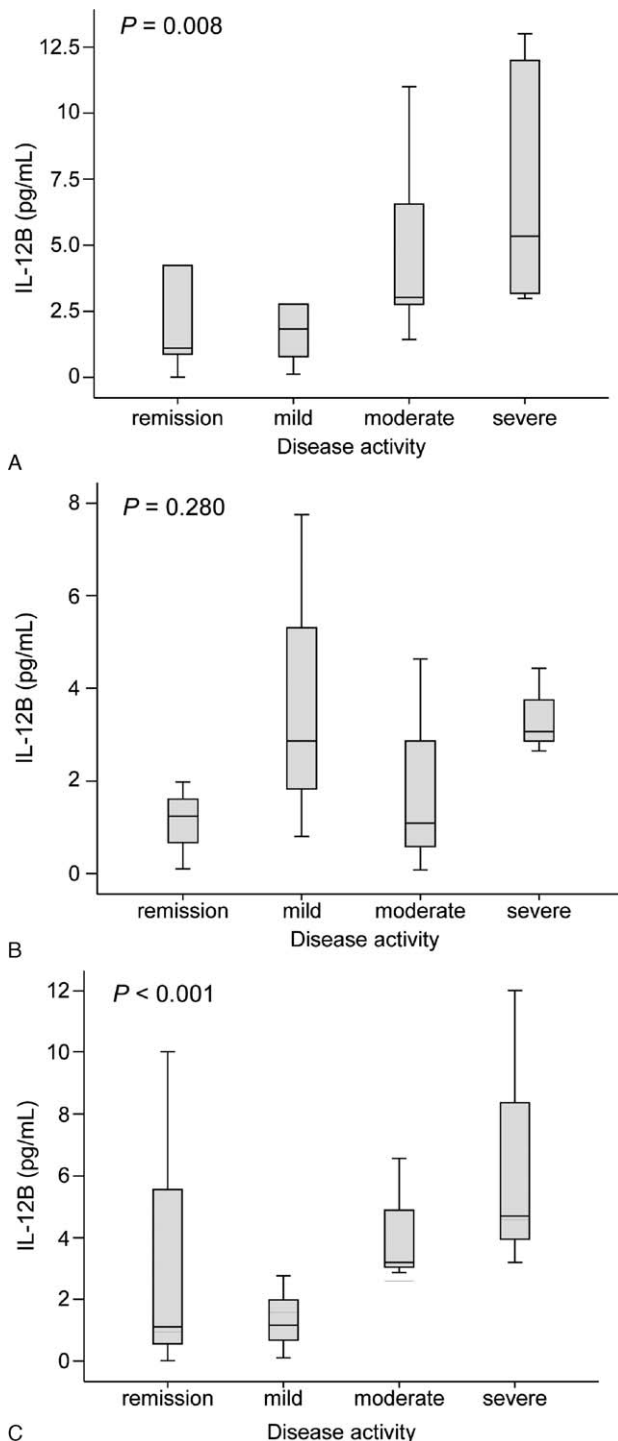
mild disease, 2.76 (range, 1.43–11.0) pg/mL among those with moderate disease, and 5.99 (range, 2.99–13.0) pg/mL among those with severe intestinal BD. A higher IL-12B level reflected greater disease activity in patients with intestinal BD ( $P = 0.008$ ).

The ESR was significantly correlated with disease activity in patients with CD ( $P = 0.045$ ) and intestinal BD ( $P = 0.005$ ), but not in those with UC ( $P = 0.470$ ). CRP levels were significantly correlated with disease activity only in patients with CD ( $P = 0.003$ ).

TABLE 2. IL-12B Levels (pg/mL) According to Clinical Disease Activity

Disease	Remission	Mild	Moderate	Severe	P Value
CD (n = 38)					
n (%)	4 (10.5)	7 (18.4)	21 (55.3)	6 (15.8)	
IL-12B, pg/mL	1.24 (0.10–1.98)	2.86 (0.80–7.75)	1.09 (0.08–4.63)	3.06 (2.65–4.43)	0.280
ESR, mm/h	45.0 (2.0–75.0)	61.0 (2.0–120.0)	65.5 (2.0–120.0)	67.0 (43.0–120.0)	0.045
CRP, mg/dL	7.8 (1.0–22.4)	10.5 (2.1–89.6)	15.3 (3.6–26.9)	21.3 (1.0–143.2)	0.003
UC (n = 32)					
n (%)	10 (31.2)	8 (25.0)	7 (21.9)	7 (21.9)	
IL-12B, pg/mL	1.10 (0.01–10.0)	1.18 (0.12–2.77)	3.19 (2.83–6.56)	4.69 (3.18–12.0)	<0.001
ESR, mm/h	12.0 (6.0–18.0)	20.0 (2.0–77.0)	16.5 (2.0–76.0)	32.0 (6.0–48.0)	0.470
CRP, mg/dL	0.6 (0.4–21.0)	3.5 (2.7–4.4)	2.7 (0.4–25.0)	10.1 (4.4–33.3)	0.584
BD (n = 32)					
n (%)	7 (21.9)	10 (31.2)	9 (28.1)	6 (18.8)	
IL-12B (pg/mL)	1.11 (0.88–4.24)	2.49 (0.79–9.0)	2.76 (1.43–11.0)	5.99 (2.99–13.0)	0.008
ESR, mm/h	13.0 (0.9–69.0)	31.0 (13.0–49.0)	40.5 (0.4–81.0)	79.0 (1.8–109.0)	0.005
CRP, mg/dL	0.6 (0.1–1.1)	5.4 (0.3–30.9)	4.1 (2.0–92.7)	5.4 (0.3–74.2)	0.431

Variables are expressed as the median (range) or n (%) as appropriate.  
BD = Behçet's disease; CD = Crohn's disease; UC = ulcerative colitis.



**FIGURE 1.** The distribution of serum interleukin (IL)-12B levels according to disease activity. A, IL-12B levels in patients with Crohn's disease (CD). B, IL-12B levels in patients with ulcerative colitis (UC). C, IL-12B levels in patients with intestinal Behçet's disease (BD).

### Levels of IL-12B and Endoscopic Disease Activity

The associations of IL-12B levels with endoscopic disease activity were analyzed (Table 3). These results were comparable to those regarding clinical disease activity. IL-12B levels

were significantly correlated with endoscopic disease activity among patients with UC and intestinal BD, but not among those with CD. In patients with CD, IL-12B levels were 1.05 (range, 0.10–2.84) pg/mL among those in remission, 1.97 (range, 1.0–7.75) pg/mL among those with mild disease, 1.24 (range, 0.08–5.66) pg/mL among those with moderate disease, and 3.46 (range, 2.65–7.05) pg/mL among those with severe CD ( $P = 0.605$ ). However, IL-12B increased with increasing endoscopic disease activity in patients with UC as follows: 1.73 (range, 0.01–2.57) pg/mL among those in remission, 1.97 (range, 1.48–3.99) pg/mL among those with mild disease, 2.88 (range, 2.04–4.32) pg/mL among those with moderate disease, and 4.50 (range, 1.44–12.0) pg/mL among those with severe UC ( $P = 0.002$ ). Regarding intestinal BD, serum IL-12B levels were 0.80 (range, 0.88–5.0) pg/mL among patients in remission, 2.71 (range, 0.79–7.47) pg/mL among those with mild disease, 2.80 (range, 1.25–5.92) pg/mL among those with moderate disease, and 6.20 (range, 4.24–13.0) pg/mL among those with severe disease ( $P = 0.001$ ).

CRP levels were significantly correlated with endoscopic disease activity in patients with CD ( $P < 0.001$ ) and intestinal BD ( $P = 0.016$ ). No statistical significance was observed regarding the association of the ESR with endoscopic disease activity in any disease group (see Supplementary STARD Flow Diagram, <http://links.lww.com/MD/B56>).

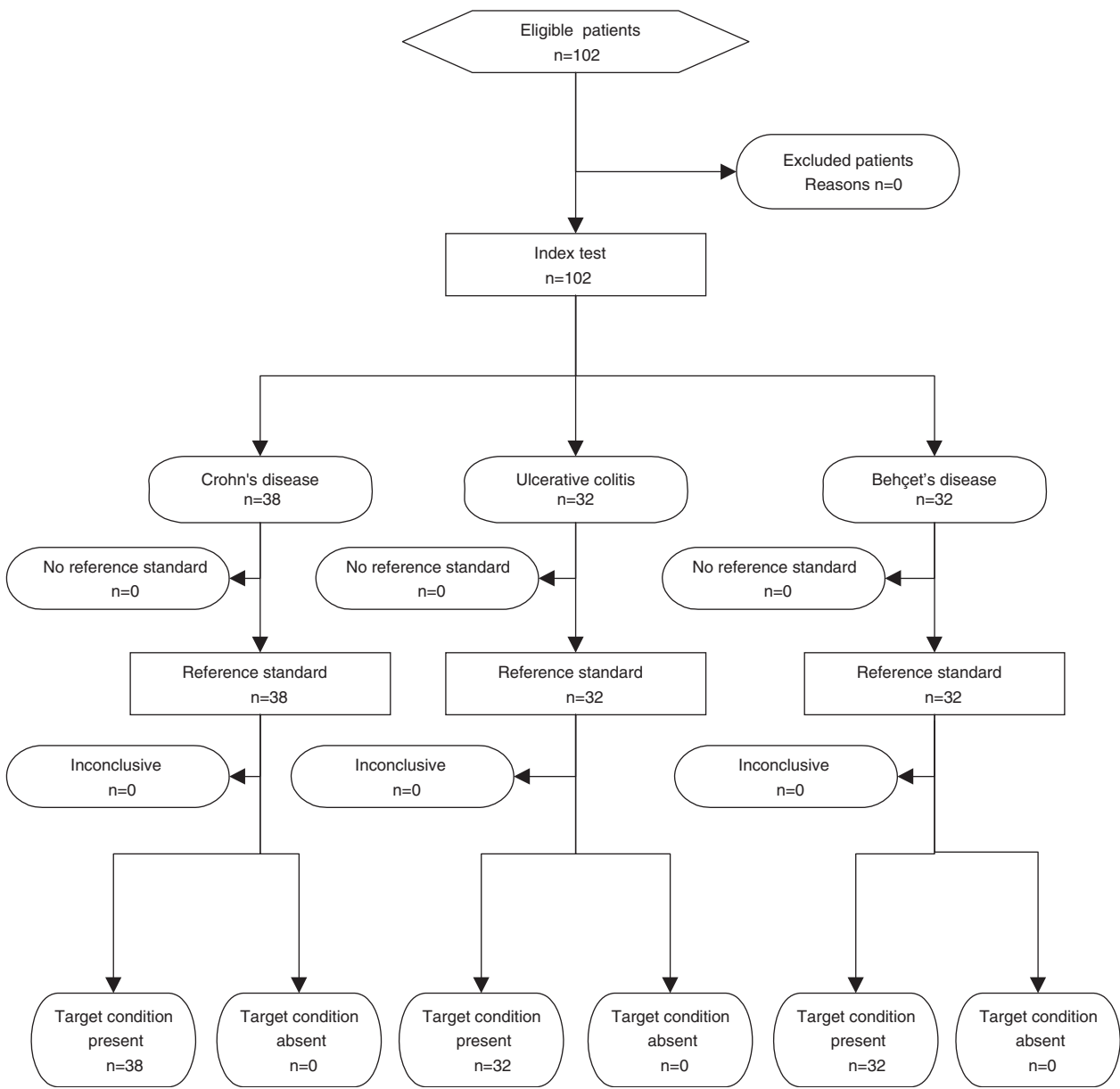
### DISCUSSION

At present, there are limited reports regarding the mechanism by which *IL12B* may functionally affect IBD susceptibility in humans. *IL12B* encodes the IL12 p40 subunit, which is common to the heterodimeric cytokines IL-12 and IL-23.<sup>17</sup> IL-12 plays a role in the development of Th1 immune responses<sup>18,19</sup> and IL-23 participates in the maintenance of Th17 cells.<sup>20</sup> These cytokines are known to be involved in the pathogenesis of various chronic inflammatory diseases.<sup>21–25</sup> Regarding IBD, several experimental studies revealed that IL-12 and IL-23 are involved in the pathogenesis of IBD.<sup>26,27</sup> Clinical studies also reported a favorable clinical response in patients with CD who were treated with ustekinumab, a human monoclonal antibody-binding IL-12/23 p40.<sup>28</sup> Furthermore, previous studies identified a significant association of genetic variants in *IL12B* with CD.<sup>6,29,30</sup>

In the present study, we focused on serum IL-12B levels as biomarkers to assess disease activity in patients with IBD. Our results uncovered that patients with IBD had higher IL-12B levels than controls. Additionally, concentrations of IL-12B were associated with clinical and endoscopic disease activity in patients with UC and intestinal BD, but not in those with CD. The result of our study might be explained by some factors. First, clinical activity indices are poorly correlated with mucosal inflammation especially for the patients with CD. In a recent large cross-sectional study, patient-reported clinical activity indices are only modest predictors of mucosal inflammation in UC and do not predict mucosal inflammation in CD.<sup>31</sup> For this reason, another study used fecal calprotectin correlated closest with Simple Endoscopic Score for Crohn's disease (SES-CD), followed by CRP, blood leukocytes, and the CDAI.<sup>16</sup> Second, this phenomenon was also noted in the other noninvasive biomarkers for IBD. A previous study identified increased levels of CRP were noted in nearly 100% of patient with CD and approximately 50% of those with UC.<sup>32</sup> However, the reason for the higher rates of increased levels of CRP in patients with CD compared with UC is still unknown.



General example



Additionally, the efficacy of therapeutic blocking agent for IL-12/23 p40 seems to be inconclusive. To date, two monoclonal antibodies neutralizing the p40 chain (ustekinumab and briakinumab) and hence blocking both IL-12 and IL-23 activity, have been developed.<sup>33</sup> The research has been reported mainly on the CD, some of them had a poor result and the others were with satisfaction. Using a novel subcutaneous dosing schedule, ustekinumab was successful in improving clinical, laboratory, and endoscopic markers of disease activity in patients with severe, refractory CD.<sup>34</sup> In a population of moderate–severe CD patients refractory to one or more prior TNF antagonists, ustekinumab induced clinical response and remission.<sup>35</sup> In

contrast, the briakinumab study in CD failed to meet its primary outcome with low remission rates.<sup>36</sup> By week 12, clinical remission rates were 11% at placebo, 29% at briakinumab 400 mg, and 22% at briakinumab 800 mg without significance.<sup>36</sup> For UC patients, ustekinumab showed good effect in phase 2 trials.<sup>37</sup> However, few studies have been conducted in UC. In our data, IL-12B reflected disease activity in UC, but not in CD. Thus, further large-scale studies are needed in the understanding of the mechanism-associated serum IL-12B and disease activity in IBD, especially UC.

To date, accurate diagnoses, assessments of disease severity, and predictions of the disease course represent challenges

**TABLE 3.** IL-12B Levels (pg/mL) According to Endoscopic Disease Activity

Disease	Remission	Mild	Moderate	Severe	P Value
CD (n = 38)					
n (%)	14 (36.8)	12 (31.6)	10 (26.3)	2 (5.3)	
IL-12B, pg/mL	1.05 (0.10–2.84)	1.97 (1.0–7.75)	1.24 (0.08–5.66)	3.46 (2.65–7.05)	0.605
ESR, mm/h	51.0 (2.0–120.0)	61.0 (2.0–120.0)	64.7 (40.0–88.0)	72.0 (65.0–120.0)	0.080
CRP, mg/dL	4.8 (1.0–15.4)	10.5 (4.0–89.6)	10.8 (8.0–143.2)	22.8 (44.0–120.0)	<0.001
UC (n = 32)					
n (%)	12 (37.5)	8 (25.0)	7 (21.9)	5 (15.6)	
IL-12B, pg/mL	1.73 (0.01–2.57)	1.97 (1.48–3.99)	2.88 (2.04–4.32)	4.50 (1.44–12.0)	0.002
ESR, mm/h	16.0 (2.0–30.0)	36.0 (2.0–50.0)	51.0 (33.0–69.0)	42.0 (4.0–77.0)	0.060
CRP, mg/dL	4.8 (0.4–8.4)	1.9 (1.0–3.8)	2.9 (2.4–5.6)	8.9 (6.5–33.3)	0.248
BD (n = 32)					
n (%)	10 (31.3)	11 (34.4)	7 (21.9)	4 (12.5)	
IL-12B, pg/mL	0.80 (0.88–5.00)	2.71 (0.79–7.47)	2.80 (1.25–5.92)	6.20 (4.24–13.0)	0.001
ESR, mm/h	4.5 (0.4–6.0)	52.0 (0.9–77.0)	45.8 (15.0–48.0)	74.6 (15.0–109.0)	0.525
CRP, mg/dL	2.8 (0.1–3.5)	2.7 (1.4–47.5)	4.0 (1.0–92.7)	4.8 (4.8–70.6)	0.016

Variables are expressed as median (range) or n (%) as appropriate.

BD = Behçet's disease, CD = Crohn's disease, CRP = C-reactive protein, ESR = erythrocyte sedimentation rate, IL-12B = interleukin-12B, UC = ulcerative colitis.

for physicians in the management of patients with IBD. Studies on IBD biomarkers have focused on markers related to genetic predisposition, disease type, inflammation or disease activity, drug-metabolism, and neoplastic transformation.<sup>38</sup> However, problems remain to be solved such as invasiveness, cost, and the need for mucosa or tissue. Currently available noninvasive biomarkers applied in the clinic are CRP, ESR, perinuclear neutrophil antibodies, antisaccharomyces cerevisiae antibody, and fecal calprotectin.<sup>38–42</sup>

In terms of serologic biomarkers, CRP is considered one of the most important proteins in acute inflammation.<sup>43</sup> In healthy individuals, levels of CRP, which is secreted by hepatocytes, remain at low levels in circulation, but its levels remarkably increase, reaching up to 350 to 400 mg/L, in the presence of acute inflammation induced by IL-6, tumor necrosis factor- $\alpha$ , or IL-1 $\beta$ .<sup>38</sup> A CRP level of 10 to 40 mg/L indicates chronic inflammation or viral infection. The half-life of CRP is extremely short, and CRP levels increase rapidly and decrease sharply in the presence of acute inflammation. CRP responds differently in patients with UC and CD. CD, but not UC, is significantly correlated with CRP levels.<sup>44,45</sup> In our study, CRP exhibited good correlations with both clinical and endoscopic disease activity in patients with CD. However, there were no significant correlations with disease activity in patients with UC or intestinal BD.

As another acute phase reactant, the ESR denotes the migration speed of red blood cells in plasma. It varies with the concentration of plasma and size of erythrocytes. The ESR is significantly altered in patients with anemia, globalism, or Mediterranean anemia.<sup>46,47</sup> The times to peak and decline are delayed for ESR compared with those of CRP. In this study, the ESR displayed significant correlations with clinical disease activity in patients with CD and intestinal BD, but not in those with UC. However, the ESR was not significantly associated with endoscopic disease activity in any disease group. Taken together, this study illustrated the limitation of the ESR and CRP levels in reflecting the disease activity of IBD, especially UC and intestinal BD. In our study, the significant associations of IL-12B levels with clinical and endoscopic disease activity in patients with UC and intestinal BD suggested that IL-12B could

be a potential biomarker for assessing these diseases. In conclusion, the complementary use of biomarkers is believed to be more helpful than the use of any individual biomarker. IL-12B is expected to be more specific for IBD, but IL-12B measurements take longer to complete than those of CRP levels or the ESR.

This study has several limitations. First, our study population was relatively small despite some positive results in the association of IL-12B with IBD disease activity. In addition, several studies reported different results. In previous papers on BD, IL-12 levels was significantly higher in active BD compared with healthy controls.<sup>48,49</sup> UC patients also expressed higher levels of IL-12 than normal individuals.<sup>50</sup> In contrast, Yan et al reported that IL-12 levels exhibited no statistical differences between patients with UC and controls.<sup>50</sup> In another study, serum IL-12B levels in patients with active BD were compared to those in controls. No significant difference was noted between IL-12B levels in patients with BD.<sup>51</sup> To confirm the convincing association of IL-12B levels with IBD disease activity, further prospective studies are needed with a larger number of patients with IBD across various ethnicities. Second, follow-up IL-12B levels were not measured. Therefore, there is a limitation in assessing the association between the changes of IL-12B levels and disease progression or regression. Despite these limitations, our results suggested that IL-12B may be an adjunctive new biomarker that can be measured in a manner convenient for both patients and physicians.

In summary, this study first attempted to identify the correlation of serum IL-12B levels with disease activity in patients with IBD. Patients with IBD had higher serum IL-12B levels than healthy controls. Serum IL-12B levels were associated with clinical and endoscopic disease activity, especially in patients with UC and intestinal BD, suggesting its possible use as a biomarker for assessing these diseases.

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