Faecal but not serum calprotectin levels look promising in predicting active disease in Behçet's syndrome patients with gastrointestinal involvement

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ABSTRACT

Objective. *The faecal calprotectin (FC) test is widely used as a non-invasive method for identifying intestinal inflammation. A recent study suggested FC may help to diagnose gastrointestinal involvement of Behçet's syndrome (GIBS). We aimed to determine whether FC helps to distinguish active from inactive intestinal involvement in GIBS.*

Methods. *We tried to contact 70 GIBS patients registered in our tertiary multidisciplinary clinic. We prospectively collected faecal specimens and serum from 39 GIBS patients who gave informed consent assessing calprotectin and CRP levels followed by a colonoscopy. We included 47 Crohn's disease (CD) patients as controls. Active disease was defined as having ulcer/s on colonoscopy. We filled the Disease Activity Index for Intestinal Behçet's Disease (DAIBD) and Crohn's Disease Activity Index (CDAI). The cut-off for positive FC was defined as ≥150 µg/g.*

Results. *Ulcers were detected in 12/39 GIBS patients. Sensitivity and specificity of the FC test for active disease was 91.7 (95%CI:61.5-99.8) and 74.1% (95%CI:53.7-88.9). Median FC and CRP levels and DAIBD scores were higher among patients with ulcers, whereas serum calprotectin and CDAI scores were not. A negative FC test was the only significant predictor of remission (OR:37.04, 95%CI:2.4-561.6; p=0.009) on multivariate analysis. Among CD patients, 16/25 active patients and 3/22 patients in endoscopic remission had a positive FC test (OR:11, 95%CI:11-49).*

Conclusion. *FC, but not serum calprotectin seems to be a useful non-invasive tool for assessing disease activity in GIBS. Whether the presence of oral ulcers can cause false positive results remains to be studied.*

Introduction

Behçet's syndrome (BS) is a multisystem vasculitis with oral and genital ulcerations, skin lesions, uveitis, arterial, venous, central nervous system and gastrointestinal involvement (1, 2). Gastrointestinal involvement is one of the most serious manifestations of BS. Mesenteric vessel involvement is thought to result in intestinal ischaemia and eventually infarction, while neutrophilic phlebitis is considered to cause intestinal ulcerations (3). Male and female patients seem to be equally affected in gastrointestinal involvement of Behçet's syndrome (GIBS) (4) and GIBS usually begins during the third and fourth decades of life. GIBS is much more frequent in the Far East compared to other countries (5). It is not clear whether differences in diagnostic criteria used in the Far East that include gastrointestinal involvement may be responsible for some of this difference (6).

Calprotectin is a calcium-and zinc binding protein complex and comprises about 60% of the cytosol of a neutrophil. It can also be found in monocytes and macrophages and released extracellularly after the initiation of innate immune response (7). Thus, it may be measured in anywhere such as serum, faeces, synovial fluid, urine and saliva where inflammation occurs (8). Among these, faecal calprotectin (FC) is one of the most frequently studied. It is helpful in the assessment of intestinal inflammation, especially in differentiating inflammatory bowel disease (IBD) from functional disorders, as well as assessing disease activity in IBD (9, 10). It has several advantages over endoscopic evaluation and traditional inflammatory markers, including C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR). First, it is a very simple

test that can be measured by enzyme-linked immunosorbent assay (ELISA). Second, when compared to assessment of gastrointestinal symptoms, it is more objective. Third, non-invasive sampling makes it more feasible than colonoscopy. Moreover, its high sensitivity and specificity to detect intestinal inflammation in stool samples stored for up to 7 days at room temperature are further advantages (9). Calprotectin has also been evaluated as a serum marker. The study of infliximab discontinuation in Crohn's disease patients in stable remission on combined therapy with immunosuppressors (STORI) found a moderate correlation between serum calprotectin (SC) and Crohn's disease activity index (CDAI), a weak correlation between FC and SC, and no correlation between SC and endoscopic activity in IBD patients (11).

GIBS shows clinical and endoscopic similarities with Crohn's disease (CD) (12). The utility of FC in diagnosing intestinal involvement among patients with BS have been investigated in a Korean study with promising results (13). However, we are not aware of studies addressing whether FC helps to distinguish active GIBS patients from those in endoscopic remission. In this context a non-invasive method rather than an endoscopic evaluation would obviously be more desirable while clinical indices and acute phase reactants do not perform sufficiently well for this purpose. In addition, in BS a weak correlation between Disease Activity Index for Intestinal Behçet's Disease (DAIBD) and endoscopic severity was shown (14) and traditional inflammatory markers such as CRP and ESR may increase due to the presence of other BS manifestations (15). Finally, little is known about the presence of ongoing endoscopic activity in GIBS patients without gastrointestinal symptoms.

In this study, we aimed to: (1) determine whether FC and SC levels are useful to assess disease activity in BS patients with gastrointestinal involvement, (2) look at their correlations with other activity parameters such as DAIBD (16) and CDAI (17) scores (Table I), and CRP levels (3) identify the predictors of endoscopic remission, and (4) assess the presence of ongoing endoscopic activity in asymptomatic GIBS patients.

Table I. Disease Activity Index for Intestinal Behçet's Disease (DAIBD) and Crohn's Disease Activity Index (CDAI).

DAIBD		CDAI	
Item	Score	Item	Weight
General well-being for 1 week		General well-being rating over 7 days	X7
Well	0	Well=0	
Fair	10	Fair=1	
Poor	20	Poor=2	
Very poor	30	Very poor=3	
Terrible	49	Terrible=4	
Fever		Antidiarrheal use	X30
< 38°C	0	No=0	
≥38°C	10	Yes=1	
Extraintestinal manifestations*	See footnotes	Extraintestinal manifestations£	X20
Abdominal pain in 1 week		Abdominal pain rating over 7 days	X5
None	0	None=0	
Mild	20	Mild=1	
Moderate	40	Moderate=2	
Severe	80	Severe=3	
Abdominal mass		Abdominal mass	X10
None	0	None=0	
Palpable mass	10	Questionable=2	
Abdominal tenderness		Definite=5	
None	0	Haematocrit (HT)	X6
Mildly tender	10	47-HT for males	
Moderately/severely tender	20	42-HT for females	
Intestinal complications&	10 per item	Bodyweight	X1
No. of liquid stools in 1 week		1-weight/standard weightx100	
0	0	No. of liquid/very soft stools each day for 7 days	X2
1–7	10		
8–21	20		
22–35	30		
≥ 36	40		

*Oral ulcer, genital ulcer, eye lesion, skin lesion, or arthralgia (5 for each), vascular or central nervous system involvement (15 for each).
&Fistula, perforation, abscess, or intestinal obstruction. £Arthritis/arthralgia, iritis/uveitis, erythema nodosum/pyoderma gangrenozum, aphthous ulcers, anal fissures, fistulas, perianal abscess, fever (>37.8°C) (1 per finding).

Materials and methods

We have around 10,000 recorded patients with BS at the Behçet's Syndrome Research Centre, Cerrahpasa Medical Faculty, Istanbul University, Turkey. Among them, there were 70 GIBS patients. Between December 2015 and October 2016, we invited all GIBS patients who were under follow-up for a colonoscopy regardless of having gastrointestinal (GI) symptoms. We prospectively collected faecal and serum samples before colonoscopy from patients who gave an informed consent. In our gastroenterology department, CD patients are routinely assessed for disease activity with FC, serum CRP levels and CDAI. We retrospectively reviewed the charts of CD patients and identified all patients who had a FC test before colonoscopy during the year 2016 year and included them as controls. Among these 22 were active patients with ulcers on colonoscopy and 25 were inactive according to colonoscopy.

Colonoscopies of BS patients were performed by the same gastroenterologist (IH), masked to patients' clinical and laboratory complaints and findings. Active disease was defined as having ulcer/s on colonoscopy. A patient with any GI symptom regardless of endoscopic and laboratory findings was defined as symptomatic. The type of involvement was defined as described previously (6). Clinical activity was assessed with CDAI and DAIBD. The cut-off for positivity in DAIBD score is ≥19. The cut-off for a positive FC test

was defined as ≥150μg/g. We repeated the FC test in patients with a positive test even if they were in remission for GI involvement.

The stool samples were collected before starting bowel preparation. Patients were also questioned regarding non-steroidal anti-inflammatory drug (NSAID) use that could increase FC levels (18) and is associated with intestinal ulcers (19). Stool samples were stored at +4°C for no longer than 3 days. FC levels were measured by Bühlmann Calprotectin ELISA test.

Venous blood samples were drawn between 8–10 a.m. after an overnight fast (10-12 h). Serum aliquots were immediately frozen and stored at -80°C until further analysis. Levels of SC were assayed by an ELISA kit (Human CALP (Calprotectin) ELISA kit, E-EL-H2357, Elabscience Biotechnology Co., Ltd, USA). Intra- and inter-coefficient of variability were 7.4% and 9.5%, respectively. Levels of CRP were assayed by an ELISA kit (Human CRP (C-reactive protein) ELISA Kit, E-EL-H0043, Elabscience Biotechnology Co., Ltd, USA). Intra- and inter- coefficient of variability were 6.8% and 8.3%, respectively.

Statistical analyses were performed using SPSS 20.0. Data were represented as median and interquartile range (IQR). Sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), positive likelihood ratio (PLR) and negative likelihood ratio (NLR) of the FC test for active disease were estimated by 2×2 tables. Comparison of a continuous variable between active and inactive patients was done with a Mann-Whitney test. Chi-square test was used for comparison of categorical variables. Correlation of variables was assessed by Pearson or Spearman correlation coefficient. Univariate associations between remission and other variables were assessed using the chi-square or Fisher's exact test. All factors with *p*-values less than 0.25 in the univariate analysis were entered into the logistic regression analysis to determine the independent predictors of remission. Odds ratios (ORs) with their 95% confidence intervals (95% CIs) were given as a measure of association. Results were considered to be significant at the 0.05 level. STARD checklist was used for accurate reporting of diagnostic accuracy (20). This study was conducted in accordance with the Declaration of Helsinki and was approved by the ethics committee of Cerrahpasa Medical Faculty, Istanbul University (345995/2015). The study was supported by Istanbul University, Scientific Research Projects Coordination Unit (Project no: 57420). The full protocol of the study can be obtained from the Ethical Committee of Cerrahpasa Medical School.

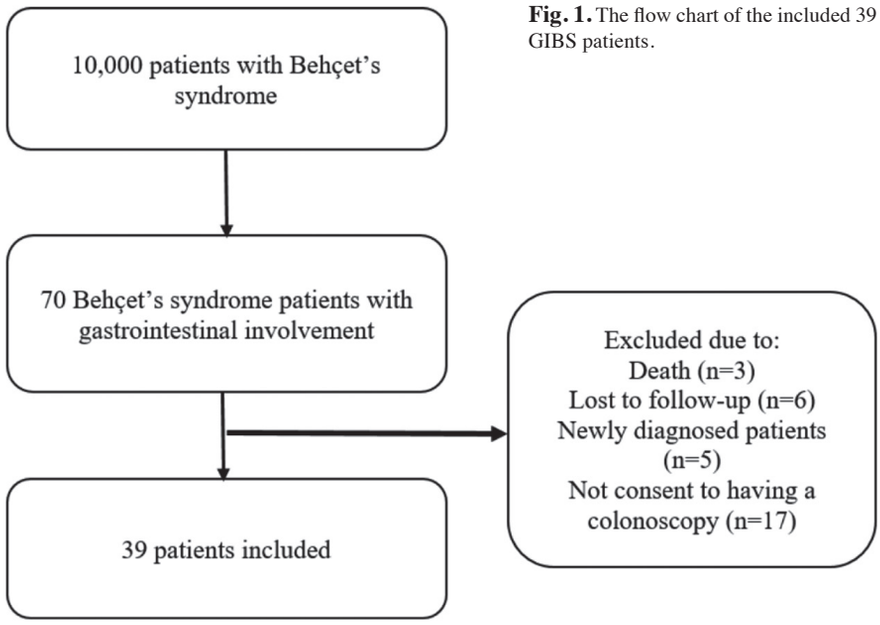

Fig. 1. The flow chart of the included 39 GIBS patients.

Table II. Demographic characteristics and other BS manifestations of 39 GIBS patients.

	GIBS patients (n=39)
Men/women (n)	19/20
Mean age ± SD (years)	42.7 ± 10.2
Mean age at diagnosis of BS ± SD (years)	28.8 ± 11.2
Mean age at diagnosis of GIBS ± SD (years)	36 ± 10.3
Juvenile onset BS (n, %)	5 (13)
Oral ulcers (n, %)	39 (100)
Genital ulcers (n, %)	34 (87)
Positive pathergy reaction (n, %)	23 (59)
Papulopustular lesions (n, %)	27 (69)
Erythema nodosum (n, %)	17 (44)
Arthritis (n, %)	19 (49)
Uveitis (n, %)	6 (15)
Deep vein thrombosis (n, %)	5 (13)
Superficial thrombophlebitis (n, %)	4 (10)
Pulmonary artery thrombosis (n, %)	0
Neurologic parenchymal involvement (n, %)	0
Dural sinus thrombosis (n, %)	1 (2)

BS: Behçet's syndrome; GIBS: gastrointestinal involvement of Behçet's syndrome.

Results

Between February 2015 and September 2016, we tried to contact 70 GIBS patients registered in our multidisciplinary clinic. Among them, 3 had died, 6 were lost to follow-up, and 5 were newly diagnosed. We asked the remaining 56 GIBS patients to have a colonoscopy. Of these, 39 patients agreed. After obtaining informed consent, we also collected faecal and serum specimens before starting bowel preparation from these patients (Fig. 1). Demographic characteristics and BS manifestations of these patients are shown in Table II. Only 1 patient with polycythemia vera was us-

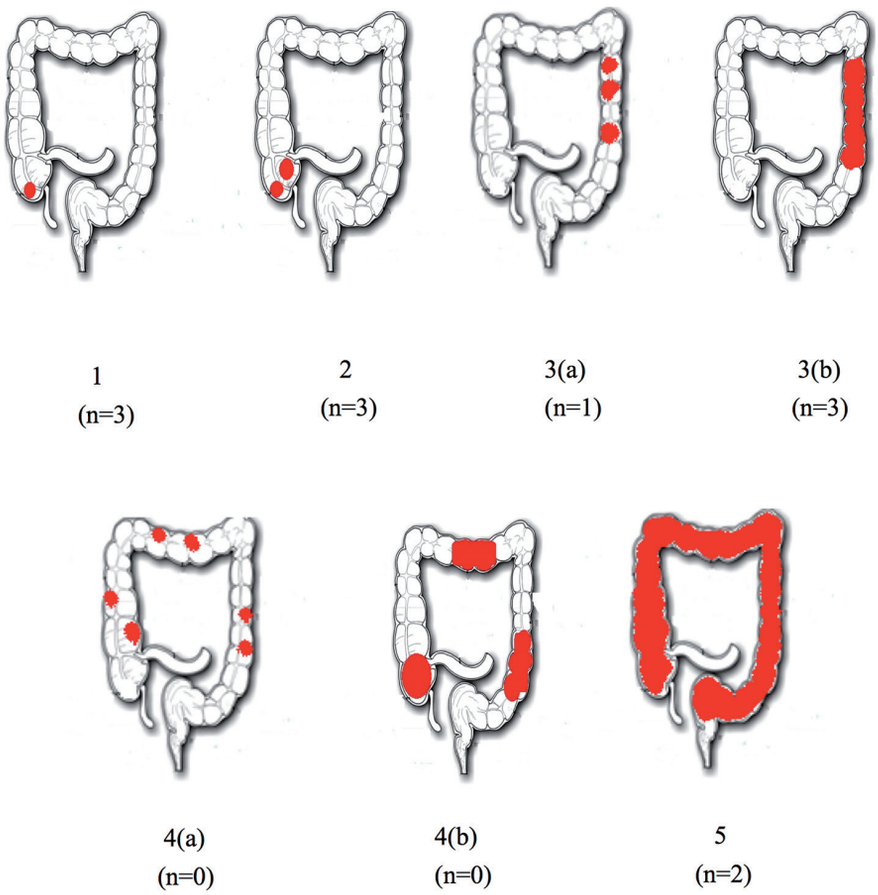

Fig. 2. The types and the frequencies of the extent of endoscopic ulcers.
1) Focal single; 2) Focal multiple; 3) Segmental, (a) multiple and (b) diffuse; 4) Multisegmental (a) multiple and (b) diffuse; 5) Pancolitis.

ing aspirin because of her haematological condition. The remaining patients did not have a history of NSAIDs use.

Twelve (31%) of 39 GIBS patients had ulcer/s on colonoscopy. Among these 12 patients, 4 (33%) had no GI symptoms. Two of them were on maintenance therapy for GI involvement and the remaining 2 were off treatment for 2 years. Six (22.2%) of 27 GIBS patients in endoscopic remission had GI symptoms.

Colonoscopic findings in active GIBS patients

Among the 12 GIBS patients who had ulcer/s (active patients), the type of involvement was focal single, focal multiple and segmental diffuse ulceration in 3 patients each. Pancolitis was observed in 2 patients and segmental multiple ulceration in 1 patient (Fig. 2). The most frequent localisation of involvement was the colonic region in 6 (50%) patients, followed by ileocaecal region in 5 (42%) and ileocolonic in 1 (8%). Among 5 patients with ileocaecal involvement, 3 had lesions on ileocaecal region and 2 had lesions in ileum. Round/oval shaped ulcers varying from 1.5 cm to 4.5 cm were the most frequent lesions, observed in 5 patients. Among them, ulcerative-type lesions and volcano shaped ulcers were accompanied by round/oval shaped ulcers in 1 patient each. Ulcerative-type lesions that were characterised by loss of vascular appearance, mucosal erythema and friability and superficial ulcerations were detected in 3 patients. However, none of them had pseudopolyps which is a typical colonoscopic finding of currently active or non-active colonic inflammation. The remaining 4 patients had apthous ulcers, 1 with accompanying linear ulcers (Fig. 3).

Faecal calprotectin

FC was positive in 11/12 (92%) GIBS patients with ulcers and in 7/27 (26%) patients without ulcers (OR: 31.4, 95% CI: 3.4 to 290). The sensitivity and specificity of the FC test for active disease was 91.7% (95%CI: 61.5-99.8) and 74.1% (95%CI: 53.7-88.9). The PPV, NPV, PLR and NLR were 61.1% (95%CI: 44.8-75), 95.2% (95%CI: 75.1-99.2), 3.54 (95%CI 1.83-6.84) and 0.11 (0.02-0.74). Median FC levels of 12 active patients were significantly higher than 27 GIBS patients in remission (301.5 μg/g (IQR: 190-1519) *vs.* 41μg/g (IQR: 30-174); $p<0.001$) (Table III). Among the 12 active patients, 8 patients with GI complaints had a higher median FC level than 4 asymptomatic patients, however, this difference was not statistically significant (638 μg/g (IQR: 239-1800) *vs.* 201.5

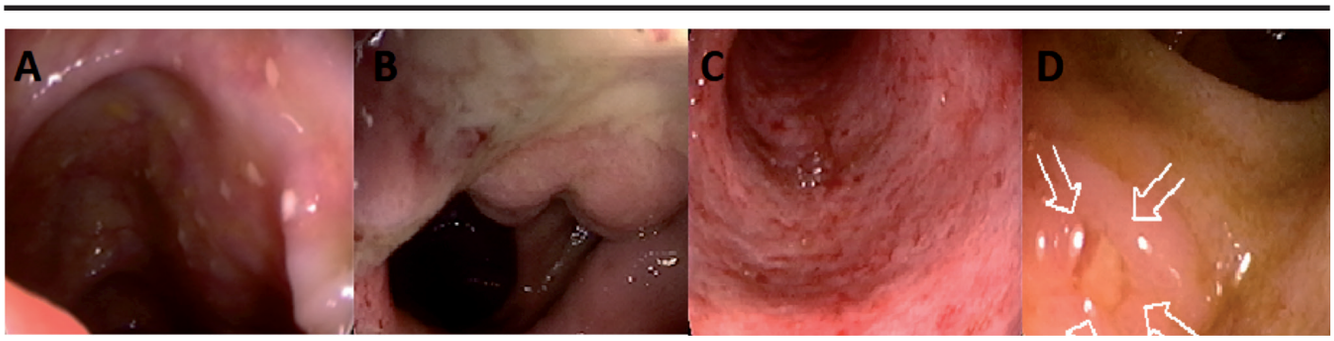

Fig. 3. The types of endoscopic ulcers. (**A**) Aphthous ulcers; (**B**) Round/oval shaped large ulcer; (**C**) Ulcerative-type lesions; (**D**) Oval volcano shaped ulcer.

μg/g (IQR: 131.5-300); *p*=0.073). Among the GIBS patients in remission, median FC levels were significantly higher in 6 symptomatic patients than in 21 asymptomatic patients (194 μg/g (IQR: 61.25-469.75) *vs*. 30 μg/g (IQR: 30-62.5); *p*=0.008). Among the 4 active patients without GI symptoms, all had a positive FC test.

Among CD patients, 16/25 active patients and 3/22 patients in remission had FC level of higher than 150 μg/g (OR: 11, 95%CI: 11 to 49) (Fig. 4).

Among the 7 GIBS patients who had high FC levels despite having no ulcers on colonoscopy, 3 had active oral aphthous lesions. When FC was repeated after the resolution of oral ulcers, the test result was negative. Two had received high dose prednisolone right after faecal sample collection which may have caused the ulcers to heal by the time colonoscopy was performed. One patient had polycythemia vera with trisomy 8 and was receiving aspirin. Finally, one patient had concomitant macrophage activation syndrome and after the resolution of macrophage activation syndrome FC test was repeated and found negative. We were not able to repeat FC test in the 2 patients who had received prednisolone and in the patient with polycythemia vera. We also do not know whether our patients had ulcers in more proximal segments of the intestine.

Table III. Faecal calprotectin, serum calprotectin, CRP and activity indices of active patients and patients in remission.

	Active GIBS (n=12)	GIBS in remission (n=27)	*p*-value
Men/women	8/4	11/16	0.17
Mean±SD age, years	47.3 ± 6.1	40.7 ± 11	0.02
Symptomatic patients, n (%)	**8 (67)**	**6 (22)**	**0.007**
Median (IQR) FC, μg/g	301.5 (190-1519)	41 (30-174)	<0.001
Fecal calprotectin ≥150 μg/g, n (%)	11 (92)	7 (22)	<0.001
Median (IQR) serum calprotectin, ng/mL	98.4 (123.5-46.8)	69 (52.3-106.7)	0.42
Median (IQR) serum CRP, mg/dL	6 (2.25-12)	2 (1-5)	0.034
CRP ≥5 mg/dL, n (%)	7 (58)	6 (22)	0.06
Median (IQR) CDAI scores	79 (20-194)	28 (0-90)	0.052
Median (IQR) DAIBD scores	45 (9-84)	20 (0-35)	0.024

GIBS: gastrointestinal involvement of Behçet's syndrome; CRP: C-reactive protein; CDAI: Crohn's disease activity index; DAIBD: disease activity index for intestinal Behçet's disease.

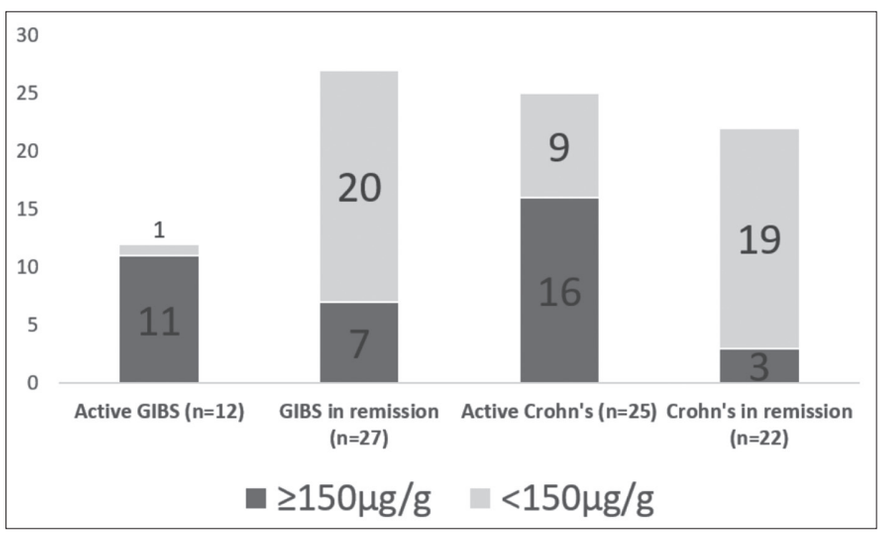

Fig. 4. Number of FC positive (≥150μg/g) test in GIBS and Crohn's disease patients who are active *vs*. in remission.

Serum calprotectin and CRP levels and intestinal activity indices

SC level was measured in 37 patients and was not different between active GIBS patients and those in remission (98.4 ng/mL (IQR: 123.5-46.8) *vs*. 69 ng/mL (IQR: 52.3-106.7); *p*=0.42). Median CRP level was significantly higher among active patients (6 mg/dL (IQR: 2.25-12) *vs*. 2 mg/dL (IQR: 1-5); *p*=0.034). However, 5/12 active patients had a normal CRP level and 6/27 patients in remission had a high CRP level (Table III).

Both CDAI (79 (20-194) *vs*. 28 (0-90); *p*=0.052) and DAIBD (45 (9-84) *vs*. 20 (0-35); *p*=0.024) scores were higher among active patients whereas this difference was significant for only DAIBD (Table III).

Correlation between activity parameters

The presence of intestinal ulcer/s were correlated with FC levels (r=0.61, *p*=0.001). There was also a correlation between the presence of intestinal ulcers and DAIBD scores (r=0.37, *p*=0.02), CDAI scores (r=0.32, *p*=0.05), CRP levels (r=0.31, *p*=0.06) and SC levels (r=0.13, *p*=0.43).

FC level was correlated with clinical indices (r=0.71, *p*<0.001 for DAIBD and r=0.64, *p*<0.001 for CDAI). There was a correlation with CRP (r=0.41, *p*=0.01) and no correlation with SC levels (r=0.02).

SC levels were not correlated with CRP levels (r= -0.16, *p*=0.33), DAIBD (r= -0.07, *p*=0.68) and CDAI scores (r= -0.004, *p*=0.98).

Predictors of remission in GIBS patients

We performed a multivariate analysis to assess the independent variables associated with remission in GIBS patients. The candidate variables we entered in the model were negative FC test, negative CRP test, being asymptomatic, and quiescent disease activity according to the DAIBD score. Multivariate analysis showed that a negative FC test was the only significant predictor of endoscopic remission (OR: 37.04, 95%CI: 2.4 to 561.6; *p*=0.09). A CRP level lower than 5 mg/L, being asymptomatic and a DAIBD score lower than 19 were not predictors of endoscopic remission.

Discussion

This study showed that: a) FC level was significantly higher among the active

GIBS patients compared to those in remission, b) FC level was ≥150 μg/g in 11 of the 12 patients with ulcers on colonoscopy indicating a good sensitivity for identifying intestinal ulcers, c) a FC level of <150 μg/g was the only predictor of endoscopic remission in a multivariate model that included acute phase reactants and clinical activity indices, d) symptomatic patients had higher FC levels even when they did not have ulcers on colonoscopy, e) 4 asymptomatic patients who had a positive FC test had intestinal ulcers suggesting that in some patients FC may be an indicator of subclinical ulcers, and f) SC levels were not useful. In the light of these findings FC assessment seems to be useful in the follow-up of GIBS patients as an indicator of endoscopic remission in asymptomatic patients with low FC levels, making control colonoscopy unnecessary. Moreover, considering the limitations of colonoscopy, cautious follow-up may be necessary for symptomatic patients with high FC levels despite normal colonoscopy. Finally, high FC levels in asymptomatic patients may be a warning sign requiring colonoscopy.

The role of FC in diagnosing GIBS has been shown in a Korean study. Kim *et al*. enrolled 44 BS patients with GI symptoms over 2 weeks (13). Of these, 25 (57%) were diagnosed as GIBS after undergoing colonoscopy. FC level was an independent predictor for a diagnosis of GIBS, but DAIBD scores and acute phase reactants were not. They proposed two cut-off values (68.89 μg/g, and 110.44 μg/g) for diagnosing GIBS. The main difference between the Korean study and the current one was the primary objectives of the studies. We looked at the role of calprotectin in the assessment of the disease activity in BS patients with gastrointestinal involvement whereas Korean study aimed to demonstrate the role of calprotectin in the diagnosis of intestinal involvement among BS patients presenting with gastrointestinal symptoms. The difference in comparator groups which were BS patients with GI symptoms due to other causes in the Korean study and BS patients with GI involvement who were in remission in our study may be responsible for the difference in the results. Moreover, their proposed cut-off values of 68.89 μg/g, and 110.44 μg/g gave sensitivity and specificity of around 80%, whereas our cut-off value of 150 μg/g that we had extrapolated from Crohn's disease showed a sensitivity of 91.7% and specificity of 74.1%. We think a high sensitivity is important here in order to avoid missing active patients. Although the primary objectives of our study and the Korean study were different, together they show that FC levels seem to be useful in both the diagnosis and activity assessment of GI involvement in BS. Our results show that FC is more useful than clinical activity indices and acute phase reactants in the assessment of disease activity and may have better ability to rule out intestinal inflammation.

One of the interesting findings of our study was that symptomatic patients had higher FC levels irrespective of having ulcers in colonoscopy. Whether this pointed to the presence of ulcers in more proximal segments of the intestine or limitations of colonoscopy remains to be verified. There were 3 previous studies assessing the role of capsule endoscopy in identifying small bowel lesions in BS patients with GI symptoms. One study demonstrated that 10/11 BS patients had small bowel lesions, 7 of them without accompanying ileocolonic lesions (21). Only 1 of them required treatment. The other study also showed lesions in all of 10 BS patients and 5 of them had no lesions in the ileocolonic region (22). Finally, a retrospective study from Japan reported that 18/19 consecutive symptomatic GIBS patients had small bowel lesions and 5 of them had no colonic or ileocecal lesions (23). It is not clear whether these findings are indicative of GIBS or nonspecific lesions. Moreover, there is no data regarding the presence of silent or subclinical ulcers among asymptomatic GIBS patients. However small bowel assessment was suggested in cases with suspected IBD and with a negative ileocolonoscopy and normal radiologic tests especially in cases with a positive FC test (24). A high FC level may point out to the necessity of capsule endoscopy or enteroscopy to investigate small bowel lesions in such patients.

FC test is thought to be specific for detecting intestinal inflammation and is helpful in differentiating inflammatory bowel diseases from non-inflammatory conditions. However, its level was also high in the other inflammatory gastrointestinal tract disorders such as NSAID enteropathy, infectious gastroenteritis, and GI malignancies (9). On the other hand, whether FC levels may be elevated during systemic inflammation is unknown. Considering that swallowed sputum during respiratory infections may influence FC results (19), it may be speculated that oral ulcerations may also increase FC levels. In our survey, among the 7 patients with a false positive FC test, repeated FC tests were negative after the resolution of systemic inflammation due to MAS in 1 and oral apthous lesions in 3. The impact of systemic inflammation and oral ulcerations on FC levels needs to be investigated further. Although it has not yet been well studied in other luminal inflammatory conditions, the change from baseline in FC may be more important in developing a strategy for follow-up of GIBS patients, rather than the value at any one-time point.

There are several studies investigating SC levels as a biomarker in various rheumatic diseases such as rheumatoid arthritis, spondyloarthropathies, psoriatic arthritis, Sjögren's syndrome, systemic lupus erythematosus, systemic sclerosis, adult onset Still disease, and vasculitides. SC levels have been found be correlated with clinical activity indices and/or acute phase reactants in all of these rheumatic diseases except for spondyloarthropathies (25). Only one study focused on SC levels in BS and found no correlation with BS disease activity (26) However, SC levels may be associated with some BS manifestations and this single study with a small number of patients was not be able to look at this possible association that was previously observed in patients with SLE (27, 28). We also observed that SC levels were not different among active GIBS patients and patients in remission suggesting that it is not useful in monitoring disease activity in patients with GIBS different from several other inflammatory

conditions. The relatively limited systematic inflammation compared to gut inflammation may be the reason of the weak association between SC levels and disease activity in GIBS, CD and spondyloarthropathies.

Our study had several limitations. First, a small number of GIBS patients were enrolled in this study. This was due to the low prevalence of GIBS in our region (5). Second, we only assessed the endoscopic but not histological remission. Although this is a hot topic in IBD, histological remission is still not a therapeutic endpoint for clinical trials and not a concern of routine practice even for IBD patients (29).

In conclusion, FC test seems to be a promising tool to assess/rule out intestinal disease activity in GIBS patients. Control colonoscopy may not be necessary in GIBS patients with a low FC level especially in asymptomatic patients. Although a positive FC level demands caution for the presence of intestinal ulcers, whether the presence of other intraluminal BS manifestations, like oral ulcers can cause false positive results remains to be further studied.

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