Original Article

Rapid Fecal Calprotectin Testing to Assess for Endoscopic Disease Activity in Inflammatory Bowel Disease: A Diagnostic Cohort Study

Lukasz Kwapisz[1], Mahmoud Mosli[1,4], Nilesh Chande[1], Brian Yan[1], Melanie Beaton[1], Jessica Micsko[1], Pauline W. Mennill[1], William Barnett[1], Kevin Bax[2], Terry Ponich[1], John Howard[1,2], Anthony Tirolese[3], Robert Lannigan[3], James Gregor[1]

[1]Departments of Medicine, [2]Pediatrics and [3]Microbiology, London Health Sciences Centre, University of Western Ontario, London, Ontario, Canada, [4]Department of Medicine, King Abdulaziz University, Jeddah, Kingdom of Saudi Arabia

Address for correspondence:
Dr. Mahmoud Mosli, Department of Medicine, King Abdulaziz University Hospital, King Abdulaziz University, Jeddah, Kingdom of Saudi Arabia.
E-mail: Mahmoud.Mosli@Robartsinc.com

ABSTRACT

Background and Aim: With increasing numbers of patients diagnosed with inflammatory bowel disease (IBD), it is important to identify noninvasive methods of detecting disease activity. The aim of this study is to examine the diagnostic accuracy of fecal rapid calprotectin (FC) testing in the detection of endoscopically active IBD. **Patients and Methods:** All consecutive patients presenting to outpatient clinics with lower gastrointestinal symptoms were prospectively recruited. Patients provided FC samples. Sensitivity (Sn), specificity (Sp), positive predictive value (PPV), and negative predictive value (NPV) for FC were calculated. Receiver–operator characteristics (ROC) curve was used to identify the ideal FC cutoff that predicts endoscopic disease activity. Correlation between FC and endoscopic disease activity, disease location, and C-reactive protein (CRP) levels were measured. **Results:** One hundred and twenty-six patients, of whom 52% were females, were included in the final analysis with a mean age of 44.4 ± 16.7 years. Comparing FC to endoscopic findings, the following results were calculated: A cutoff point of 100 μg/g showed Sn = 83%, Sp = 67%, PPV = 65%, and NPV = 85%; and 200 μg/g showed Sn = 66%, Sp = 82%, PPV = 73%, and NPV = 77%. Based on ROC curve, the best FC cutoff point to predict endoscopic disease activity was 140 μg/g. Using this reference, FC levels strongly correlated with colorectal, ileocolonic, and ileal disease and predicted endoscopic activity. **Conclusions:** FC is an accurate test when used as an initial screening tool for patients suspected of having active IBD. Given its noninvasive nature, it may prove to reduce the need for colonoscopy and be an added tool in the management of IBD.

Key Words: Crohn's, diagnostic accuracy, inflammatory bowel disease, ulcerative colitis

Received: 12.05.2015, Accepted: 02.09.2015
How to cite this article: Kwapisz L, Mosli M, Chande N, Yan B, Beaton M, Micsko J, *et al*. Rapid fecal calprotectin testing to assess for endoscopic disease activity in inflammatory bowel disease: A diagnostic cohort study. Saudi J Gastroenterol 2015;21:360-6.

Patients presenting to gastroenterologists with abdominal pain and/or diarrhea are often suspected of having either inflammatory bowel disease (IBD) or irritable bowel syndrome (IBS). Classically, IBD is a chronic disease that has a relapsing and remitting pattern and its two main entities include Crohn's disease (CD) and ulcerative colitis (UC). Symptoms typically associated with IBD include diarrhea, rectal bleeding, abdominal pain, and weight loss. Over the past several decades there has been a dramatic rise in the incidence of IBD worldwide.[1,2] Similarly, there has been a parallel increase in the rates of IBS.[3,4] Compared with IBS, untreated IBD can be associated with poor outcome. Therefore, distinguishing IBD from IBS is necessary.

Traditionally, both noninvasive tests such as C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) as

Access this article online
Quick Response Code:
Website: www.saudijgastro.com
DOI: 10.4103/1319-3767.170948

well as invasive procedures such as ileocolonoscopy and flexible sigmoidoscopy have been used to diagnose and evaluate IBD for disease activity. The drawback to serological markers, such as CRP and ESR are that they lack specificity for intestinal inflammation and can be elevated in systemic inflammatory states. Conversely, the disadvantages to colonoscopy are rooted in the nature of being invasive, uncomfortable for patients, and being associated with risk of adverse events. As such, there has been a growing need for simple, cost-effective, and noninvasive tests to detect intestinal inflammation and aid in differentiating IBD from IBS.

Fecal calprotectin (FC) is a biomarker that has demonstrated promise in its specificity for intestinal inflammation.[5] Calprotectin is a calcium- and zinc-binding protein that constitutes nearly 60% of neutrophil cytosolic protein.[6] Levels of calprotectin therefore indicate the presence of neutrophils and other immune cells released from damaged intestinal mucosa.[7] Calprotectin's major functions include both antibacterial and antifungal activity, inhibition of metalloproteinases, and induction of apoptosis.[8] Two large meta-analyeis have showed that FC is a very useful tool in discriminating between IBD and IBS.[5,9] The pooled sensitivity and specificity of FC was found to be as high as 93% and 96%, respectively. However, in those studies FC levels were measured using an enzyme-linked immunosorbent assay (ELISA) method and endoscopy was not always used as reference standard. More recently, less cumbersome and more efficient rapid point of care tests have been developed, which can reliably provide results within 15 min.[10,11] A recent study showed that rapid FC testing has a positive correlation both to clinical and endoscopic severity in IBD.[12] This advantage may provide clinicians with the means to apply on site "personalized medicine" care in an efficient way. The calprotectin immunoassay by Quantum Blue (Buhlmann Laboratories, Schönenbuch, Switzerland) is designed to provide valuable information about the status of mucosal inflammation; can aid in predicting relapses and can detect calprotectin within a wide range of 100–1800 μg/g. These criteria in theory can provide even higher sensitivity in cases where inflammation is expected such as in patients with an established diagnosis of IBD.

The aim of our study is to evaluate the diagnostic accuracy of rapid point of care FC testing and correlate its results with endoscopic disease activity, disease location, and CRP as an aid in evaluating and establishing the diagnosis of IBD.

MATERIALS AND METHODS

Study population

We performed a prospective cohort study involving 130 consecutive adult patients, who were recruited between January 2013 and June 2014, presenting to specialized IBD outpatient gastroenterology clinics at London Health Sciences Centre (LHSC; University Hospital and Victoria Hospital) and St. Joseph's Hospital with signs and symptoms suggestive of active IBD. Diagnosis of IBD was based on typical clinical, endoscopic, and radiologic criteria supported by histological evidence of the disease. Inclusion criteria included age between 18 and 85 years as well as symptoms suggestive of IBD or a previously established diagnosis of IBD with symptoms of activity. All patients were investigated with either colonoscopy or flexible sigmoidoscopy within 4 weeks of their presentation as part of their diagnostic workup. Exclusion criteria included inability to perform endoscopy within the allocated time frame, a positive stool test for *Clostridium difficile* toxin or inability to provide a stool sample for analysis.

Study methods

Stool samples were collected from each patient (1:1) and subsequently analyzed for FC levels. Results were then compared with reports of the patient's endoscopic evaluation. Endoscopic assessments were performed by 10 board certified gastroenterologists, who were blinded to the results of the index test, as were the study personnel who received the FC results blinded to the endoscopic reports. The reference standard of endoscopic disease activity was used to determine the overall diagnostic accuracy of the test. Endoscopic evidence of active inflammation was noted when there was a mucosal break, exudate, or ulcerations present in keeping with traditional scoring systems such as the Mayo score for UC and simple endoscopic scoring for CD (SES-CD).[13,14] In the absence of any mucosal abnormalities in the colon and terminal ileum; the endoscopic assessment was reported as normal. Data on demographics, medications, clinical parameters, and laboratory investigations were simultaneously collected.

Outcomes

The primary objective of this study was to assess the diagnostic accuracy of high-range rapid FC immunoassay in the diagnosis and assessment of IBD when compared with endoscopic activity. Secondary objectives included identifying the optimal FC cutoff point and using it to correlate FC levels with endoscopic disease activity, disease location, and CRP levels.

Methods of fecal calprotectin determination

All patients provided a stool sample within four weeks of their scheduled endoscopic evaluation. Less than 1 g of native stool was required. Samples could be kept refrigerated at 2°C—8°C for up to 6 days. If the samples could not be analyzed by the sixth day, they were kept frozen at −20°C and subsequently thawed when ready for measurement. All samples were analyzed by a high-range FC quantitative point of care test (FC-QPOCT), which is manufactured

by Bühlmann, Quantum Blue (Buhlmann Laboratories, Schönenbuch, Switzerland).[15]

The QPOCT is a quantitative lateral flow assay, which with the help of a reader system provides results ranging from 100 to 1800 μg/g. The procedure consisted of three steps. First, samples were extracted. Next, 1:150 Chase Buffer solution was used to dilute the stool samples, followed by vortex and centrifuge. Lastly, 80 μL of diluted stool sample were loaded onto a cassette of a Quantum Blue Reader and a result was provided within 15 min. Two microbiologists who were appropriately trained were assigned to perform the FC measurements, while being blinded for endoscopic results and patients' clinical status.

Statistical analysis and sample size calculation

Descriptive statistics were expressed as means (±standard deviations (SD)) for continuous variables and as proportions for categorical variables. One-way ANOVA test was used to compare means. Sample size calculation was based on the premise that FC is a highly sensitive test for the evaluation of patients with suspected IBD. Assuming ileocolonoscopy is the gold standard with a sensitivity of 100%, and assuming a type 1 error of 0.05 and 80% power we estimated that 122 patients will need to undergo both tests to determine a sensitivity of fecal calprotectin of 95%.

Sensitivity (Sn), specificity (Sp), positive predictive value (PPV), and negative predictive values (NPV) were calculated using 100 and 200 μg/g as cutoff points and endoscopic activity as reference standard.

Simple linear regression was used to measure correlation. Pearson's correlation coefficient (r) and Spearman's rank correlation coefficient (rho) were used to express correlation between FC and CRP levels and disease location, respectively. Receiver operator characteristic (ROC) curve analysis was used to identify the FC-QPOCT and CRP points that best predict endoscopic disease activity for IBD. Simple and multiple logistic regression analyses were then used to characterize the relationship between abnormal FC levels, based on the cutoff point identified by ROC curve analysis, and disease location (rectal, colonic, colorectal, ileal and ileocolonic) and endoscopic activity (active versus inactive). Associations were expressed as odds ratios (OR). StataCorp (TX, USA) statistical software, version 12.0 was used for statistical analysis and a two-sided 5% significance level was used for all statistical inferences. Precision of estimates was measured using 95% confidence intervals (95% CI).

Ethical considerations

The Western University Research Ethics Board for Health Sciences Research Involving Human Subjects (HSREB) provided approval (#102975) for the conduction of this clinical study. All recruited patients provided written informed consent for participation in the study.

RESULTS

Patients' characteristics

Baseline characteristics of the study population are summarized in Table 1. Of the 170 patients who were eligible and offered enrollment, 130 provided stool samples for FC analysis. During the course of the study, four patients were not able to undergo endoscopy (colonoscopy or flexible sigmoidoscopy) and the final analysis was based on 126 patients. The mean age was 44.4 ± 16.7 and 52% were females. Exactly 50% of enrolled patients had known IBD prior to their endoscopy, whereas the other 50% had symptoms suggestive of IBD but had not yet been diagnosed. Endoscopic changes consistent with IBD were found in 53 (41%) patients, with 11 (8%) patients being newly diagnosed. Lastly, patients enrolled in the study presented to gastroenterologists with a variety of symptoms related to IBD including sole diarrhea at 22%, rectal bleeding at 21%, abdominal pain at 18%, and a mixture of multiple symptoms present 39% of the time.

Primary end points

In this particular study, a newer version of a point of care immunoassay for FC was used to determine the diagnostic accuracy of the test. According to the manufacturer, values below 100 μg/g are indicative of either a noninflammatory state or mild inflammation in the gut. The results in our study for FC levels below 100 μg/g generated Sn = 83%, Sp = 67%, PPV = 65%, and NPV = 85%. Conversely, using FC values greater than 200 μg/g as a cutoff point, which is intended by the manufacturer to indicate active organic disease with inflammation in the gastrointestinal tract, resulted in Sn = 66%, Sp = 82%, PPV = 73%, and NPV = 77% [Table 2].

Table 1: Baseline demographics and clinical characteristics (*N*=126)

Demographics	
Age in years (mean±SD)	44.4±16.7
Male gender, No. (%)	62 (48)
Female gender, No. (%)	68 (52)
Known IBD, No. (%)	65 (50)
No known IBD, No. (%)	65 (50)
Newly diagnosed IBD, No. (%)	11 (8.5)
Presenting symptoms, No. (%)	
Diarrhea	28 (22)
Rectal bleeding	27 (21)
Abdominal pain	24 (18)
Multiple symptoms	51 (39)

*IBD: Inflammatory bowel disease, IBS: Irritable bowel syndrome

Table 2: Diagnostic accuracy results for FC; cut-off points of <100, <200, and <140 µg/g, and for CRP using cut-off points s of <5, <7, and <10 mg/L

Test and cut-off point	TP	FP	TN	FN	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	LR+	LR−
FC (mcg/g)										
<100	44	24	49	9	83	67	65	85	1	-
<140	41	19	54	12	77	74	68	82	2.8	0.3
<200	35	13	60	18	66	82	73	77	3.7	0.4
CRP (mg/dL)										
<5	17	10	26	22	44	72	63	54	1.4	0.8
<7	15	9	27	24	38	75	63	53	1.5	0.8
<10	10	8	28	29	26	78	56	49	1	-

TP: True positive, FP: False positive, TN: True negative, FN: False negative, PPV: Positive predictive value, NPV: Negative predictive value, LR: Positive likelihood ratio, LR: Negative likelihood ratio, FC: Fecal calprotectin, CRP: C-reactive protein

Secondary end points

For our cohort of patients, 60% had levels of CRP measured. Compared with active inflammation observed through endoscopy, CRP values (using a cutoff of 5 mg/dL) in this study showed Sn = 44%, Sp = 72%, PPV = 63%, and NPV = 54% [Table 2].

Location of inflammation was also recorded and correlated to FC levels. Overall, 29 patients had active disease limited to the colon or rectum (55%), 16 patients had activity localized to the ileum (30%), and 8 patients had ileocolonic involvement (15%). Within each location, Sn was calculated and found to be 90% for the colon/rectum, 88% for ileocolonic disease, and 69% for the ileum. Also, consistently higher FC levels were observed for active disease that had at least partial colonic involvement (whether colon and rectum or ileocolonic). When only the terminal ileum was involved, a much lower sensitivity was observed as well as lower overall FC levels. There was a statistically significant difference between groups as determined by one-way ANOVA (F (4,48) = 5.04, P =0.0018) [Table 3].

To better characterize the correlation between FC and CRP, simple linear regression was performed. FC, though weakly, nevertheless significantly positively correlated with CRP levels (r = 0.017, 95% CI: 0.006–0.03, P = 0.003) [Figure 1] and more strongly positively correlated with disease location (rho = 0.5191, 95% CI: 0.006–0.03, $P \leq 0.00001$).

ROC curve analysis identified 140 µg/g as the optimal FC cutoff point to predict IBD endoscopic disease activity (area under the curve (AUC) =0.81, 95% CI: 0.73–0.87) with Sn = 77%, Sp = 73%, positive likelihood ratio (LR+) = 2.8 and negative likelihood ratio (LR−) =0.3 [Figure 2]. Similarly, the optimal cutoff point for CRP was identified as 4 mg/dL (area under the curve (AUC) =0.584, 95% CI: 0.46–0.69) with Sn = 60%, Sp = 60%, positive likelihood ratio (LR+) = 1.5 and negative likelihood ratio (LR−) = 0.7.

Table 3: Disease location and its impact on FC sensitivity

Location	Active disease	% Sensitivity (FC <100 µg/g)	Mean FC if active disease present (±SD)
Rectum	5	60	366 µg/g (±570)
Colon	13	100	1020 µg/g (±696)
Colorectum	11	91	1211 µg/g (±685)
Ileum	16	69	341 µg/g (±358)
Ileocolon	8	88	1026 µg/g (±701)

FC: Fecal calprotectin, SD: Standard deviation. A statistically significant difference between groups was determined by one-way ANOVA testing (F (4,48)=5.04, P=0.0018)

Using this cutoff point, FC results were dichotomized into normal and abnormal. Simple and multiple logistic regression analyses were performed. Abnormal FC was highly predictive of endoscopic disease activity on simple (OR = 9.7, 95% CI: 4.24–22.25; $P \leq 0.0001$) and multiple (OR = 11.3, 95% CI: 2.68–47.68; P = 0.001) logistic regressions (adjusted for disease location). FC was also predictive of disease location (OR = 1.99, 95% CI: 1.43–2.77; $P \leq 0.0001$). Similar analysis was performed to identify disease locations that predicted an abnormal FC test result. A significant positive association was observed with colorectal (OR = 27.14, 95% CI: 3.27–225.15; P = 0.002), ileocolonic (OR = 19, 95% CI: 2.21–163.79; $P \leq 0.007$) and ileal disease (OR = 3.49, 95% CI: 1.15–10.56; $P \leq 0.027$), and colonic involvement by itself perfectly predicted an abnormal FC level as no patient with active colonic disease had a normal FC result. There was a significantly negative association between FC and rectal disease (OR = 0.085, 95% CI: 0.035–0.205; $P \leq 0.0001$) [Figure 3].

DISCUSSION

In this study, we examine the diagnostic accuracy with a rapid FC point of care assay (FC-QPOCT) and correlate its levels to disease locations, CRP levels, and endoscopic disease activity. We used a new POC test that has high-range

detection capability but is also designed to provide results within 15 min, allowing a much more efficient and useful method compared with prior arduous ELISA methods. The results we report are consistent with it being a highly accurate method of FC measurement in relation to a well-established gold standard, which has not been used as comparator in many previously reported studies.[16-18] A recent study by Lobaton *et al*. reported, using a cutoff point of 160 μg/g, an Sn of 64.9% and an Sp of 83.9% with the similar FC assay and demonstrated a very strong correlation with an ELISA-based FC assays (Intra-class correlation coefficient (ICC = 0.904, 95% CI: 0.864-0.932; P = 0.001).[18] Since the literature supports that disease activity, based on clinical criteria, does not highly correlate with endoscopic disease activity in IBD patients, especially with CD[19] we elected not to use this as a clinical endpoint.

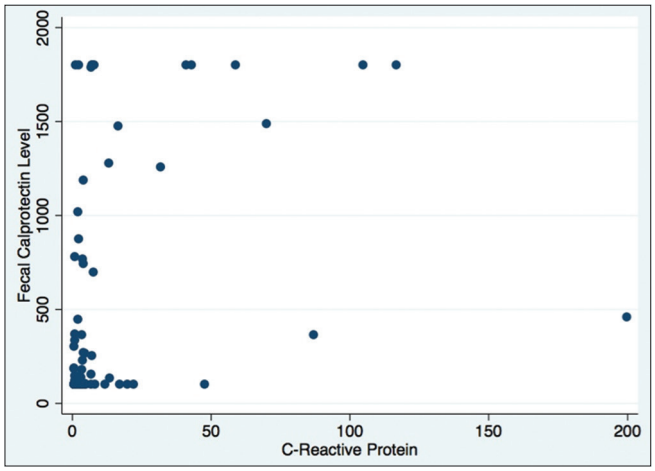

Figure 1: A scatter plot correlating fecal calprotectin with C-reactive protein levels

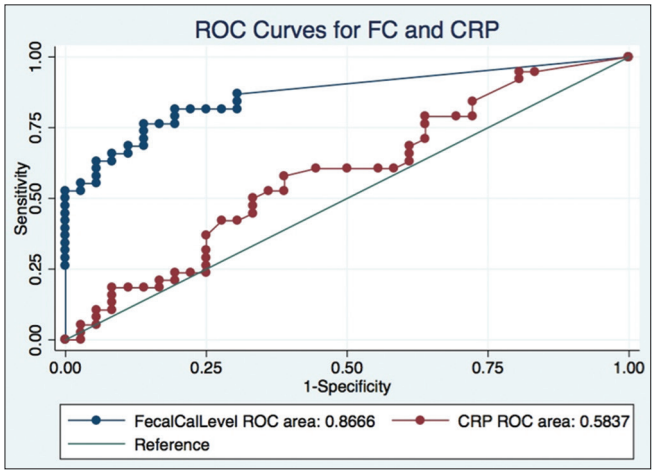

Figure 2: Receiver operator characteristic curve analysis identified 140 μg/g and 4 mg/dL, as the optimal cutoff points for fecal calprotectin and C-reactive protein, respectively

With a growing need to find new, reliable, and sensitive biomarkers in the evaluation of IBD, FC has proven to be quite valuable in discriminating between IBD and IBS.[20] The population of patients we included in this study is representative of the majority of patients with IBD, with a prominent bi-modal age group distribution. Two evenly matched groups of patients were enrolled into the study with either signs or symptoms suggestive of IBD or established diagnosis of IBD with symptom activity. This adds to the generizability of our results. It was found that for the lower cutoff value of 100 μg/g, Sn of 83% and NPV of 85% were observed. For the higher cutoff of 200 μg/g, a higher overall Sp of 82% and PPV of 73% were noted. These figures are in line with previously reported diagnostic accuracy studies for ELISA-based FC assays.[21-23] The high NPV and low LR − we report, can be useful clinical characteristics of FC to consider when it is used to triage IBD patients presenting with lower GI symptoms for colonoscopy, as negative results would be highly suggestive of inactive disease.

Further subanalysis demonstrated that areas of active disease played a prominent role in the diagnostic accuracy of FC. For colonic involvement, the overall sensitivity of FC was much better. With sole involvement of the ileum or the rectum, FC levels proved to be much less sensitive. Furthermore, there were six patients who had evidence of pancolitis and their average FC levels were the highest observed at an

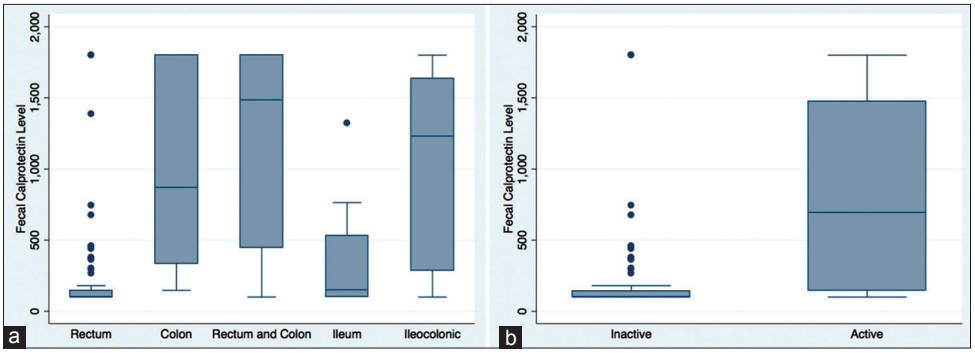

Figure 3: Box plots correlating fecal calprotectin levels with (a) disease location, and (b) endoscopic findings

average of 1710. It can be surmised from our study, that the high-range immunoassay used for FC is potentially much more valuable and reliable for colonic IBD and cannot be relied on in patients with distal "rectal" UC.

Classically, CRP has been considered a poor marker of IBD activity with reported sensitivity estimates as low as 50%.[24] In our study, only 60% of patients received blood work, which included CRP levels prior to their endoscopy. We recognize the limitation around this analysis, but still found Sn of 44%, Sp of 72%, NPV of 54%, and PPV of 63% and that CRP poorly correlated with FC-QPOCT. One can see FC's superiority over CRP despite the limited data.

The ideal cutoff point for FC tests has been a subject of debate and results have varied widely.[25-27] Our results, which are based on a properly powered study sample, suggest that 140 μg/g most accurately predicts endoscopic disease activity and this correlated well with endoscopic disease activity and disease location. The limitation to this analysis is that the FC-QPOCT does not report results below 100 or above 1800 μg/g and therefore cutoff points outside that range could not be assessed.

Lastly, it is important to mention that we did not have strict exclusion criteria and acknowledge that there are a variety of conditions that can artificially increase FC levels as studies show that benign conditions such as menstrual bleeding and epistaxis as well as diseases such as bacterial enteritis/colitis, eosinophilic esophagitis, microscopic colitis, peptic ulcer disease, colorectal cancer, polyps, diverticulitis, and nonsteroidal anti-inflammatory drugs can all induce an increase in FC readings.[28-33] Overall, 23 patients in our study were found to have polyps on endoscopy. Of those patients, 64% had elevated FC levels without endoscopic evidence of IBD. As a result, one can see that the presence of polyps can certainly affect the interpretation of FC levels. Furthermore, some important clinical confounders such as disease severity were not controlled for in our analyses and we acknowledge these limitations. Nevertheless, rapid point of care FC assays are highly accurate in detecting endoscopic disease activity for IBD, especially colonic disease, and can be used to triage patients for colonoscopy in an effort to preserve resources and improve patient care.

CONCLUSIONS AND RECOMMENDATIONS

Although FC has shown to be a very useful addition to the diagnosis of IBD, many questions still remain. An optimum cutoff has not yet been determined in literature and even though FC has shown tremendous promise in discerning between IBD and IBS, its exact future remains unclear. FC's destiny may lie as a biomarker that is used to trend response to therapy in disease states or be a simple tool that primary care doctors along with gastroenterologists can use to screen for IBD prior to endoscopy. In our study, it is important to note that FC levels were much more accurate when disease activity was found in the colon, this factor suggests that its use may be more beneficial in UC and Crohn's colitis versus noncolonic CD. Further studies are certainly on the horizon for FC and in time it may prove to be a very valuable tool for daily use in the investigation of numerous gastrointestinal diseases. Based on our results, we recommend using FC-QPOCT to screen patients suspected of having IBD using a cutoff point of 140 μg/g to discriminate between active and inactive disease.

Acknowledgment

We would like to acknowledge and thank all the nurses and hospital staff who assisted us in acquiring the study samples.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

REFERENCES

1. Benchimol EI, Guttmann A, Griffiths AM, Rabeneck L, Mack DR, Brill H, *et al*. Increasing incidence of paediatric inflammatory bowel disease in Ontario, Canada: Evidence from health administrative data. Gut 2009;58:1490-7.
2. Gismera CS, Aladrén BS. Inflammatory bowel diseases: A disease(s) of modern times? Is incidence still increasing? World J Gastroenterol 2008;14:5491-8.
3. Quigley EM, Abdel-Hamid H, Barbara G, Bhatia SJ, Boeckxstaens G, De Giorgio R, *et al*. A global perspective on irritable bowel syndrome: A consensus statement of the World Gastroenterology Organisation Summit Task Force on irritable bowel syndrome. J Clin Gastroenterol 2012;46:356-66.
4. Canavan C, West J, Card T. The epidemiology of irritable bowel syndrome. Clin Epidemiol 2014;6:71-80.
5. van Rheenen PF, Van de Vijver E, Fidler V. Faecal calprotectin for screening of patients with suspected inflammatory bowel disease: Diagnostic meta-analysis. BMJ 2010;341:c3369.
6. Røseth AG, Fagerhol MK, Aadland E, Schjønsby H. Assessment of the neutrophil dominating protein calprotectin in feces. A methodologic study. Scand J Gastroenterol 1992;27:793-8.
7. Erbayrak M, Turkay C, Eraslan E, Cetinkaya H, Kasapoglu B, Bektas M. The role of fecal calprotectin in investigating inflammatory bowel diseases. Clinics (Sao Paulo) 2009;64:421-5.
8. Sutherland AD, Gearry RB, Frizelle FA. Review of fecal biomarkers in inflammatory bowel disease. Dis Colon Rectum 2008;51:1283-91.
9. Lin JF, Chen JM, Zuo JH, Yu A, Xiao ZJ, Deng FH, *et al*. Meta-analysis: Fecal calprotectin for assessment of inflammatory bowel disease activity. Inflamm Bowel Dis 2014;20:1407-15.
10. Coorevits L, Baert FJ, Vanpoucke HJ. Faecal calprotectin: Comparative study of the Quantum Blue rapid test and an established ELISA method. Clin Chem Lab Med 2013;51:825-31.
11. Kolho KL, Turner D, Veereman-Wauters G, Sladek M, de Ridder L, Shaoul R, *et al*. Rapid test for fecal calprotectin levels in children with

Crohn's disease. J Pediatr Gastroenterol Nutr 2012;55:436-9.
12. Tursi A, Elisei W, Giorgetti G, Picchio M, Brandimarte G. Rapid fecal calprotectin correlates with clinical and endoscopic severity of inflammatory bowel diseases. Scand J Gastroenterol 2013;48:1359-60.
13. Schroeder KW, Tremaine WJ, Ilstrup DM. Coated oral 5-aminosalcylic acid therapy for mildly to moderately active ulcerative colitis. A randomized study. N Engl J Med 1987;317:1625-9.
14. Daperno M, D'Haens G, Van Assche G, Baert F, Bulois P, Maunoury V, *et al*. Development and validation of a new, simplified endoscopic activity score for Crohn's disease: The SES-CD. Gastrointest Endosc 2004;60:505-12.
15. Sydora MJ, Sydora BC, Fedorak RN. Validation of a point-of-care desk top device to quantitate fecal calprotectin and distinguish inflammatory bowel disease from irritable bowel syndrome. J Crohns Colitis 2012;6:207-14.
16. Schoepfer AM, Trummler M, Seeholzer P, Criblez DH, Seibold F. Accuracy of four fecal assays in the diagnosis of colitis. Dis Colon Rectum 2007;50:1697-706.
17. Guardiola J, Lobatón T, Rodríguez-Alonso L, Ruiz-Cerulla A, Arajol C, Loayza C, *et al*. Fecal level of calprotectin identifies histologic inflammation in patients with ulcerative colitis in clinical and endoscopic remission. Clin Gastroenterol Hepatol 2014;12:1865-70.
18. Lobatón T, López-García A, Rodríguez-Moranta F, Ruiz A, Rodríguez L, Guardiola J. A new rapid test for fecal calprotectin predicts endoscopic remission and postoperative recurrence in Crohn's disease. J Crohns Colitis 2013;7:e641-51.
19. Sipponen T, Nuutinen H, Turunen U, Färkkilä M. Endoscopic evaluation of Crohn's disease activity: Comparison of the CDEIS and the SES-CD. Inflamm Bowel Dis 2010;16:2131-6.
20. Waugh N, Cummins E, Royle P, Kandala NB, Shyangdan D, Arasaradnam R, *et al*. Faecal calprotectin testing for differentiating amongst inflammatory and non-inflammatory bowel diseases: Systematic review and economic evaluation. Health Technol Assess 2013;17:xv-xix, 1-211.
21. Tibble JA, Sigthorsson G, Bridger S, Fagerhol MK, Bjarnason I. Surrogate markers of intestinal inflammation are predictive of relapse in patients with inflammatory bowel disease. Gastroenterology 2000;119:15-22.
22. Gisbert JP, Bermejo F, Pérez-Calle JL, Taxonera C, Vera I, McNicholl AG, *et al*. Fecal calprotectin and lactoferrin for the prediction of inflammatory bowel disease relapse. Inflamm Bowel Dis 2009;15:1190-8.
23. Gaya DR, Lyon TD, Duncan A, Neilly JB, Han S, Howell J, *et al*. Faecal calprotectin in the assessment of Crohn's disease activity. QJM 2005;98:435-41.
24. Vermeire S, Van Assche G, Rutgeerts P. Laboratory markers in IBD: Useful, magic, or unnecessary toys? Gut 2006;55:426-31.
25. Kallel L, Ayadi I, Matri S, Fekih M, Mahmoud NB, Feki M, *et al*. Fecal calprotectin is a predictive marker of relapse in Crohn's disease involving the colon: A prospective study. Eur J Gastroenterol Hepatol 2010;22:340-5.
26. D'Haens G, Ferrante M, Vermeire S, Baert F, Noman M, Moortgat L, *et al*. Fecal calprotectin is a surrogate marker for endoscopic lesions in inflammatory bowel disease. Inflamm Bowel Dis 2012;18:2218-24.
27. Molander P, af Björkesten CG, Mustonen H, Haapamäki J, Vauhkonen M, Kolho KL, *et al*. Fecal calprotectin concentration predicts outcome in inflammatory bowel disease after induction therapy with TNFα blocking agents. Inflamm Bowel Dis 2012;18:2011-7.
28. Kopylov U, Rosenfeld G, Bressler B, Seidman E. Clinical utility of fecal biomarkers for the diagnosis and management of inflammatory bowel disease. Inflamm Bowel Dis 2014;20:742-56.
29. Wang S, Wang Z, Shi H, Heng L, Juan W, Yuan B, *et al*. Faecal calprotectin concentrations in gastrointestinal diseases. J Int Med Res 2013;41:1357-61.
30. Licata A, Randazzo C, Cappello M, Calvaruso V, Butera G, Florena AM, *et al*. Fecal calprotectin in clinical practice: A noninvasive screening tool for patients with chronic diarrhea. J Clin Gastroenterol 2012;46:504-8.
31. Pavlidis P, Chedgy FJ, Tibble JA. Diagnostic accuracy and clinical application of faecal calprotectin in adult patients presenting with gastrointestinal symptoms in primary care. Scand J Gastroenterol 2013;48:1048-54.
32. Rogler G, Aldeguer X, Kruis W, Lasson A, Mittmann U, Nally K, *et al*. Concept for a rapid point-of-care calprotectin diagnostic test for diagnosis and disease activity monitoring in patients with inflammatory bowel disease: Expert clinical opinion. J Crohns Colitis 2013;7:670-7.
33. Shastri YM, Bergis D, Povse N, Schäfer V, Shastri S, Weindel M, *et al*. Prospective Multicenter study evaluating fecal calprotectin in adult acute bacterial diarrhea. Am J Med 2008;121:1099-106.