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REVIEW

Utility of surrogate markers for the prediction of relapses in inflammatory bowel diseases

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Abstract Patients with diagnosed inflammatory bowel disease (IBD) will commonly experience a clinical relapse in spite of a prolonged therapy-induced period of clinical remission. The current methods of assessing subclinical levels of low-grade inflammation which predispose patients to relapse are not optimal when considering both cost and patient comfort. Over the past few decades, much investigation has discovered that proteins such as calprotectin that are released from inflammatory cells are capable of indicating disease activity. Along with C-reactive protein and erythrocyte sedimentation rate, calprotectin has now become part of the current methodology for assessing IBD activity. More recently, research has identified that other fecal and serum biomarkers such as lactoferrin, S100A12, GM-CSF autoantibodies, α1-antitrypsin, eosinophil-derived proteins, and cytokine concentrations have variable degrees of utility in monitoring gastrointestinal tract inflammation. In order to provide direction toward novel methods of predicting relapse in IBD, we provide an up-to-date review of these biomarkers and their potential utility in the prediction of clinical relapse, given their observed activities during various stages of clinical remission.

Keywords Monitoring · Calprotectin · GM-CSF · Lactoferrin · S100 · C-reactive protein

Introduction

Inflammatory bowel disease (IBD)—Crohn's disease (CD) and ulcerative colitis (UC)—arises from complex interactions between genetic and environmental factors, and is characterized by periods of remission and relapse managed by active therapy and disease monitoring. With the main focus of IBD treatment centered around the induction and maintenance of clinical remission through pharmacotherapy, there can still be low levels of inflammation present in the human gastrointestinal tract (GIT), leading to an increased risk of disease relapse [1]. The ability to accurately predict a relapse in IBD would enable clinicians to make alterations in pharmacotherapy and thus keep patients in remission. Currently, CD activity and UC activity are monitored through a combination of clinical indices and invasive investigations such as imaging, luminal endoscopy, and histopathology. Whilst accurate in their ability to assess IBD activity, these investigations are both costly and/or time-consuming, significant drawbacks for both clinician and patient [2]. In order to manage IBD in a less-invasive manner, there is a need for accurate biomarkers to assess disease activity, leading to the potential to predict relapse. To date, serological markers of inflammation such as C-reactive protein (CRP) and erythrocyte sedimentation rate (ESR) are commonly used, but they are nonspecific to IBD and still require a blood test for sample collection [1]. Fecal biomarkers, however, are noninvasive and generally accepted by the patient, which gives them the potential to alleviate the above challenges in monitoring IBD subclinical activity. Such fecal biomarkers

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are either released by activated inflammatory cells or are cytokines, which cross the intestinal mucosa and can be detected in fecal samples. S100A8/S100A9, better known as calprotectin, is to date the most well studied, and has been shown to differentiate IBD from other pathologies of the GIT as well as to indicate disease activity. Along with the acute-phase proteins and calprotectin, we review here other fecal and serum biomarkers of GIT inflammation, such as lactoferrin and S100A12, and assess their ability to accurately predict a relapse in IBD and thus their future role in the clinical management of CD and UC. Quantitative fecal immunochemical testing (FIT) for hemoglobin is an alternative modality that is being considered for use in IBD and is currently less expensive than fecal calprotectin tests. The test gives a numerical result for the stool human hemoglobin concentration using an antibody specific for human hemoglobin. FIT for hemoglobin has mainly been investigated as a method for colorectal cancer screening [3, 4]. However, recent studies indicate a future role for quantitative occult blood testing in disease monitoring in IBD [5–7].

Given that the aim of this review is to ascertain the predictive value of biomarkers for IBD relapse, all attempts have been made to focus solely on this aspect of relevant studies. Therefore, though clinically relevant to IBD as a whole, any studies or findings relating to remission and its tracking will not be discussed. Along these lines, studies that find no association between biomarkers and positive relapse but do find one between those biomarkers and the prediction of continued remission are discussed in minor detail.

Methods

The systematic review was performed using PubMed/MEDLINE up to January 2015. The search strategy used the following search terms alone or in combination: monitoring, biomarker, marker, surrogate, evaluation, prediction, predictor, response, responder, healing, recurrence, relapse, remission, management, efficacy, outcome, flare, calprotectin, serum, fecal, faecal, blood, lactoferrin, S100, C-reactive protein, serological, inflammatory bowel disease, Crohn's disease, ulcerative colitis. Boolean operators ("not," "and," "or") were also used in succession to narrow or widen the search. Title, abstract, and full article selection was performed independently by two reviewers (JD, JODM), with conflicts resolved by consensus adjudication. The primary outcome was to review the clinical utility of biomarkers for the prediction of relapses in IBD.

Acute-phase reactants

In normal conditions, only low levels of CRP can be detected in the blood (<1 mg/L); however, after any acute-phase event, hepatocytes can rapidly increase its production. This increase in CRP production is detectable in the bloodstream and is often correlated to the severity of the insult [8–10]. The upregulation in production often occurs within hours of the event, which, in conjunction with the relatively short half-life of CRP, makes it a valuable acute-phase reactant for the detection and monitoring of inflammation. However, the CRP responses to certain inflammatory conditions can be quite heterogeneous; whilst CD is often associated with a relatively strong CRP response, UC often gives rise to a mild or absent response [11–14].

Multiple studies dating back to the 1980s have investigated the efficacy of CRP as a biomarker specifically for relapse prediction in IBD (Table 1). Following on from previous studies identifying the possible importance of CRP in predicting the course of IBD [15, 16], Boirivant et al. [17] followed a group of CD patients over the course of 24 months, measuring the CRP and calculating the Bristol Simple Index [18] to evaluate the clinical activity of the disease at each encounter. With a mixed population of patients with active disease and those in remission on entry, they were able to identify that there was no significant relationship between the serum CRP value at entry and the clinical outcome within the first 2–6 months. Interestingly, however, they did show that the likelihood of a clinical relapse during the 12- to 24-month period following entry into the study was higher in patients with persistently high CRP levels in the year prior than in those with low CRP levels [17], a finding which has been both supported [19, 20] and refuted [21] by other studies.

In a broad cohort of patients, Kiss et al. [22] looked at whether high-sensitivity CRP (hs-CRP) had any prognostic value in predicting clinical relapse of patients during normal follow-up consultations. Using the Harvey–Bradshaw index [23] (HBI) and the European Crohn's and Colitis Organization (ECCO) guidelines [24], they prospectively assessed 260 patients over the course of 12 months in those who showed no relapse, or until the date of relapse in those who did. They found that the predictive value of hs-CRP was highly associated with the initial hs-CRP value at the time of diagnosis; hs-CRP values in patients in clinical remission predicted relapse at 3 and 12 months with reasonable accuracy in those with a high hs-CRP (>10 mg/L) at the time of diagnosis, though the predictive value was limited for those with an initially normal hs-CRP [22]. Of note, however, they also determined that, when compared to disease location, non-inflammatory disease behavior,

Table 1 Studies investigating C-reactive protein for the prediction of relapses in inflammatory bowel disease

Study	Patient population	Subjects (*n*)	Cutoff level	Sensitivity/ specificity (%)	PPV/ NPV (%)	Treatments allowed whilst in remission	Relapse definition
Kennedy et al. [27]	Adult IBD	CD: 129 UC: 108	NA	NA	NA	None	Mild: topical treatments, commencement/increase in dose of oral 5-aminosalicylate Moderate: oral corticosteroids or thiopurines Severe: admission to hospital, surgery, IV corticosteroids or anti-TNFα usage
Boirivant et al. [17]	Adult CD	25	1 mg/dL	NA	NA	None	Bristol Simple Index ≥4
Kiss et al. [22]	Adult CD	260	3 month: 10.1 mg/L 12 month: 8.8 mg/L	65/71 52/75	70/67 68/62	No restrictions	HBI >4, ΔHBI ≥3, and change in medical therapy
Koelewijn et al. [20]	Adult CD	94	15 mg/L	NA	NA	None	Symptoms, confirmed by (ileo)colonoscopy/small bowel follow through, hospitalization/initiation of corticosteroids or immunosuppressant
Bitton et al. [19]	Adult CD	101	10 mg/L	NA	NA	No restrictions	CDAI >150 with increase of ≥70 above baseline
Jurgens et al. [25]	Adult CD	268	NA	NA	NA	Infliximab	Development of clinical symptoms
Park et al. [26]	Adult CD	45	0.5 mg/dL	NA	NA	Thiopurines	CDAI ≥150
Brignola et al. [28]	Adult CD	41	NA	NA	NA	None	CDAI >150 and ΔCDAI ≥100 from entry level for 2 weeks

IBD inflammatory bowel diseases, *CD* Crohn's disease, *UC* ulcerative colitis, *PPV* positive predictive value, *NPV* negative predictive value, *CDAI* Crohn's disease activity index, *HBI* Harvey–Bradshaw Index, *NA* not available

frequency of relapses, and perianal involvement, only hs-CRP was independently associated with the probability of a clinical relapse at 3 months ($P = 0.007$), whilst both hs-CRP ($P = 0.001$) and perianal involvement ($P = 0.01$) were similarly associated with clinical relapse at 12 months in the subgroup with a high hs-CRP at the time of diagnosis [22].

Many studies have attempted to identify subsets of IBD patients in which CRP may be of particular predictive benefit. Jürgens et al. [25] studied a cohort of patients on maintenance infliximab therapy, measuring their CRP at baseline and at regular follow-up appointments over the course of 5 years. For those who suffered a clinical relapse within that time, the baseline CRP measurements were higher than in those who did not relapse ($P = 0.012$) [25]. Interestingly, they also noted that a CRP increase preceded the clinical relapse in 69 % of those patients, though the recorded values never quite reached those of the baseline [25]. Looking at a slightly different subset of IBD patients, Park et al. [26] found that in patients who were undergoing thiopurine treatment for medically or surgically induced remissions, an increased CRP level (>0.5 mg/dL) measured at the time of initial remission was an independent predictor of relapse ($P = 0.013$) [26]. More recently, Kennedy et al. [27] reported that in CD patients in stable clinical remission on long-term thiopurine therapies who were withdrawn from treatment, univariable analysis revealed a high association between the CRP measured at the time of thiopurine cessation and the risk of relapse within 12 months ($P = 0.005$), though this was not as predictive in UC patients.

Brignola et al. [28] studied a population of adult CD patients in clinical remission and who had not been on any form of drug therapy for 6 months, monitoring them at both 9- and 18-month follow-up appointments using the Crohn's disease activity index (CDAI) and a host of laboratory investigations. When dividing the patient groups according to outcome (relapsed and non-relapsed) and comparing their respective results, they found that acid alpha-1-glycoprotein, alpha-2 globulin, and ESR were significantly different to a high degree between the two groups, with CRP and alpha-1 antitrypsin differing to a less

statistically significant level. Interestingly, they derived a prognostic index based on these laboratory parameters with a threshold of discriminant power of 0.35. The accuracy of the index by the 18-month mark was 88 %, with a sensitivity and specificity of 71 and 100 %, respectively, though it performed rather less admirably at the 9-month mark (sensitivity and specificity of 62 and 94 %, respectively).

Whilst unable to effectively predict short-term relapse, a biological predictive score (BPS) developed by Consigny et al. [29] has the potential to be used in the prediction of continued remission. The patient cohort used was identical to the cohort used by Modigliani et al. [30], and had been receiving oral steroid treatment prior to entering the study at the time of weaning their dose. Whilst following up patients in remission every 6 weeks, assessments of CRP, ESR, and a range of other serum levels were used to assist in the development of the BPS. Of the 71 patients that could be included, 38 returned to active CD as defined by the CDAI. The Cox proportional hazards model was used to assess the relative risk of relapse and thus to define the predictive biomarkers to be used in their BPS. A CRP level >20 mg/L and ESR >15 mm/h were deemed predictive of relapse ($P < 0.001$) and incorporated into their derived predictive score. With the focus of this study being on the prediction of CD relapse in the 6 weeks following a patient's most recent follow up, the authors found the relative risk of such an event to be 4 (if CRP elevated) and 4.8 (ESR) when using the Cox model. These predictive capabilities were further supported in the same study when looking at CRP and ESR levels together. Taking this multivariate approach, elevation of both CRP and ESR resulted in a 9.9 times relative risk of CD relapse in the coming period compared to those with normal CRP and ESR values. The BPS derived from the above model showed similar results, and a positive score indicated an 8 times relative risk of relapse with the score sensitivity and specificity 89 and 43 %, respectively. Interestingly, the NPV of 97 % indicates that a negative score at any follow-up would give confidence in remission continuing for the next 6 weeks, but the poor PPV (15 %) would suggest that a positive score would not confidently predict relapse in the short term. Nevertheless, this simple score could be used to predict short-term maintenance remission of CD following a recent flare treated with corticosteroids.

Similarly, a risk factor panel which included two biomarkers, CRP and perinuclear antineutrophil cytoplasmic antibodies (pANCA), was identified by Arias et al. [31] and focused on the ability to predict relapse of UC. Primarily focusing on the outcome of patients recently started on infliximab therapy ($n = 285$) following a lack of response to corticosteroids or immunomodulatory agents, the authors were able to identify a linear relationship between an increasing number of panel risk factors a patient exhibited and clinical relapse of UC within 12 months ($P < 0.001$). The presence of pANCA in patient serum along with elevated CRP above baseline levels seen in remission were used as biomarkers in the risk panel. Other risk factors included hypoalbuminemia, a lack of response to infliximab therapy, and absence of mucosal healing. Twenty-five percent of patients with zero or one of these risk factors experienced clinical relapse of UC within 12 months, but this proportion increased significantly to 83 % for patients who had four or five of the risk factors on the developed predictive panel. Whilst this study was retrospective and lacked standardized criteria for UC activity, it did provide support to the notion seen in the studies of Brignola et al. [28] and Consigny et al. [29] that a biomarker such as CRP may have a role within an index, score, or panel that has the ability to predict relapse of IBD.

Calprotectin

Calprotectin is expressed in granulocytes, monocytes, epithelial cells, and neutrophils [32, 33]. It possesses certain characteristics that make it an attractive biomarker for gastrointestinal disorders; not only is its representation in the feces proportional to neutrophil influx into the gastrointestinal mucosa during inflammation, but it shows resistance to fecal degradation and stability at room temperature [34, 35]. Though the study of calprotectin in gastrointestinal disease has been rapid and widespread, its value specifically in IBD as a noninvasive biomarker for disease progression and relapse prediction has garnered much interest (Table 2).

In a landmark paper assessing the feasibility of measuring intestinal inflammation directly to predict clinical relapse in IBD patients via fecal calprotectin, Tibble et al. [36] found that in patients who had been in clinical remission for between 1 and 4 months, a fecal calprotectin level of greater than 50 mg/L had a high predictive value for clinical relapse during the following 12-month period [36]. Interestingly, though CRP and ESR had already been previously identified as biomarkers with the potential for predicting relapse [15–17, 28], this study found them to hold no significant predictive value. Following on from this, when differentiated into two separate subgroups of IBD, Costa et al. [37] reported that whilst fecal calprotectin (cutoff value of 150 μg/g) showed good predictive value for identifying CD patients who might relapse within 12 months, it had a far stronger predictive value in those with UC. These high calprotectin levels amounted to increased risks of up to twelvefold greater for relapse within the UC group, and a fourfold increase in those with CD. In line with the prior study mentioned, Costa et al. [37]

Table 2 Studies investigating fecal calprotectin for the prediction of relapses in inflammatory bowel disease

Study	Patient population	Subjects (*n*)	Cutoff level	Sensitivity/specificity (%)	PPV/NPV (%)	Treatments allowed whilst in remission	Relapse definition
Tibble et al. [35, 36]	Adult IBD	CD: 43 UC: 37	50 mg/L	90/83	NA	No restrictions	CD: CDAI >150 with increase of >100 from inclusion value UC: HBI >4 with increase of >2 from inclusion value
Costa et al. [37]	Adult IBD	CD: 38 UC: 41	150 μg/g	CD: 87/43 UC: 89/82	CD: 50/83 UC: 81/90	No restrictions	Occurrence or worsening of symptoms, accompanied by an increase in CDAI >150 or in UCAI >4, sufficient to require a change in therapy
D'Inca et al. [103]	Adult IBD	CD: 65 UC: 97	130 mg/kg	CD: 68/67 UC: NA	CD: 52/79 UC: NA	No restrictions	Worsening clinical picture with CDAI >150, an increment of more than 50 points over baseline (or 75 in resected patients), or ET scores exceeding 4 and requiring additional treatment
Wagner et al. [100]	Adult IBD	CD: 11 UC: 27	NA	NA	NA	No restrictions	CD: nonresponse defined as unchanged or increased HBI UC: nonresponse defined as a decrease in only clinical or endoscopic score or unchanged or increased clinical and/or endoscopic score
Molander et al. [50]	Adult IBD	CD: 16 UC: 33	140 μg/g 200 μg/g	79/53 83/50	NA	No restrictions	CD: HBI ≥8 or increase >3 points to at least HBI = 5 UC: partial Mayo score ≥3
Garcia-Sanchez et al.[48]	Adult IBD	CD: 66 UC: 69	CD: 200 μg/g UC: 120 μg/g	CD: 80/65 UC: 81/63	CD: 46/88 UC: 49/88	No restrictions	Worsening of symptoms accompanied by CDAI ≥150 or modified TW ≥11
Orlando et al. [64]	Adult CD	39	50 μg/g 100 μg/g 150 μg/g 200 μg/g 250 μg/g	94/25 78/45 78/55 63/75 47/85	NA	Not specified	Presence of typical CD lesions in the neoterminal ileum and/or anastomosis on endoscopy
Laharie et al. [59]	Adult CD	65	130 μg/g 250 μg/g	61/48 43/57	NA	Maintenance immunosuppressant, corticosteroids	CDAI >150
Louis et al. [60]	Adult CD	115	300 μg/g	NA	NA	No restrictions except infliximab	CDAI >250 or CDAI 150–250 with increase of 70 over two consecutive weeks
Kallel et al. [40]	Adult CD	53	340 μg/g	80/91	NA	None	CDAI >150 or increase >100 from inclusion value
Naismith et al. [46]	Adult CD	92	240 μg/g	80/74	28/97	No NSAIDs	CDAI >150
Guardiola et al. [104]	Adult UC	59	155 μg/g	78/71	54/89	No restrictions	Active histological inflammation defined as presence of neutrophils infiltrating crypt epithelial cells
Lasson et al. [65]	Adult CD	30	100 μg/g 200 μg/g 250 μg/g	85/35 54/53 46/53	50/75 47/60 43/56	No restrictions	>5 Aphthoid lesions, lesions confined to anastomosis or diffuse inflammation on endoscopy

Table 2 continued

Study	Patient population	Subjects (*n*)	Cutoff level	Sensitivity/specificity (%)	PPV/NPV (%)	Treatments allowed whilst in remission	Relapse definition
Meuwis et al. [63]	Adult CD	79	250 μg/g	NA	NA	No restrictions except infliximab	CDAI >250 or CDAI 150–250 with increase of 70 over two consecutive weeks
Sipponen et al. [39]	Pediatric IBD	CD: 26	100 μg/g	NA	40/75	5-ASA, azathioprine, or nothing	PGA score of mild, moderate, or severe disease activity
		UC: 41	570 μg/g		39/NA		
		Other: 5	855 μg/g		44/NA		
			1000 μg/g		43/NA		
Walkieqicz et al. [105]	Pediatric IBD	CD: 11	400 μg/g	NA	NA	Not specified	PGA score of mild, moderate, or severe disease activity
		UC: 21					
Diamanti et al. [45]	Pediatric IBD	CD: 32	IBD: 275 μg/g	IBD: 97/85	IBD: 85/97	No restrictions	Histologically active disease—diffuse inflammatory cell infiltrate involving crypts and lamina propria; formation of crypt abscesses in the mucosa, cryptitis, crypt distortion, decreased goblet cells, Paneth cell metaplasia
		UC: 41	CD: 462 μg/g	CD: 100/89	CD: 90/100		
			UC: 275 μg/g	UC: 94/95	UC: 94/95		
Gerasimidis et al. [38]	Pediatric CD	15	NA	NA	NA	Exclusive enteral nutrition with polymeric liquid diet	PCDAI >10

IBD inflammatory bowel diseases, *CD* Crohn's disease, *UC* ulcerative colitis, *PPV* positive predictive value, *NPV* negative predictive value, *(P)CDAI* (pediatric) Crohn's disease activity index, *UCAI* ulcerative colitis activity index, *5-ASA* 5-aminosalicylates, *HBI* Harvey–Bradshaw Index, *ET* Edwards and Truelove, *TW* Truelove Witts, *PGA* physician's global assessment, *NA* not available

found no additional predictive value for ESR and CRP when combined with calprotectin.

Following these studies, the role of calprotectin in IBD rapidly gained widespread interest, and whilst a few groups have found no clinical significance of fecal calprotectin in the prediction of relapse of IBD [38, 39], there are many that have. Kallel et al. [40] were one of the first groups to independently support the original findings of Tibble et al. [36], concluding that a fecal calprotectin cutoff level of 340 μg/g produced the greatest sum of sensitivity (80 %) and specificity (91 %) for predicting clinical relapse in patients with quiescent CD. Though it was postulated that the cutoff level was similar to that of the aforementioned study when the quantitative difference in measuring techniques was taken into account [41, 42], the still slightly higher level in this study was possibly confounded by the patient population and their tendency to have had a more severe course of CD [40]. Though the admittedly small numbers of patients in the study precluded the calculation of positive and negative predictive values (NPV) of fecal calprotectin, Walkiewicz et al. [43] ascertained that a fecal calprotectin cutoff value of 400 μg/g did have a significant association with clinical relapse within 9 months ($P = 0.03$) in asymptomatic paediatric CD patients. However, some studies have called into repute the validity of clinical indices in the evaluation of IBD activity within the pediatric population [44], so in the first study to compare the histological evaluation of IBD relapse with fecal calprotectin levels, Diamanti et al. [45] showed that a cutoff value of 275 μg/g gave the greatest sum of sensitivity (97 %) and specificity (85 %) for predicting histological relapse in the IBD group as a whole, whilst a higher cutoff of 462 μg/g yielded the highest diagnostic precision for CD relapse prediction alone. In one of the largest prospective studies to date in this field, Naismith et al. [46] looked to ascertain the predictive value of fecal calprotectin in CD patients with quiescent disease over a 12-month follow-up period. Looking at a larger cohort of relatively undifferentiated CD phenotypes, they concluded that using a fecal calprotectin cutoff value of 240 μg/g had a statistically significant predictive value of a low risk of clinical relapse within 12 months, with sensitivities and specificities of 80 and 74 %, respectively. However, a relatively low positive predictive value (PPV) and a conversely high NPV limited the usefulness of this cutoff as a tool for predicting patients in whom stable remission is expected rather than clinical relapse, a finding echoed by some studies done previously [47, 48]. Furthermore, it has been shown that consecutive fecal calprotectin measurements can predict relapse in UC patients receiving infliximab maintenance therapy [49], as well as in IBD patients in deep remission after cessation of TNFα-blocking therapy [50] already 3 months before the flare. These results indicate that the utility of fecal biomarkers is partly dependent on the clinical setting at the time of sampling. This also includes considerations regarding the within-day and intra-individual variability of fecal calprotectin levels in IBD patients. A recent study showed that the day to day variability of fecal calprotectin is low in a cohort of patients with quiescent CD, and suggests that using a single calprotectin sample is sufficient to inform therapeutic strategies [51]. However, fecal calprotectin concentrations widely vary between motions in patients with active IBD [52–54]. This suggests that a single fecal calprotectin determination should not be used as the basis for therapeutic decisions in patients with active IBD.

In general, fecal calprotectin correlates better with ileocolonic disease than with isolated ileal disease. In predicting small bowel inflammatory changes, fecal calprotectin has moderate specificity but low sensitivity [55]. A recent study showed that, even in the presence of large or very large ulcers, patients with ileal CD may not have markedly elevated fecal calprotectin levels [56]. Interestingly, a study of children with new-onset untreated CD found that median fecal calprotectin levels did not differ between children with small bowel only and those with colonic involvement [57]. More studies are needed to clarify the role of fecal biomarkers in the prediction of relapses of isolated ileal CD (reviewed in [58]).

Not all studies have shown positive results for fecal calprotectin as a predictive tool. In a prospective study looking at a cohort of patients with refractory luminal CD upon initiation of infliximab therapy and maintenance immunomodulators alone, Laharie et al. [59] were not able to identify any reliable fecal calprotectin cutoff value for the prediction of relapse over one year when measured at both the baseline and at week fourteen. Following on from this study, Louis et al. [60] observed a patient cohort of infliximab responders post cessation of therapy and recognized a conglomerate of six demographic, clinical, and laboratory elements which were able to identify a subgroup of patients who were unlikely to relapse within 2 years. Though fecal calprotectin with a cutoff value of 300 μg/g was one of these variables, this study would seem to lend support to the findings of Laharie et al. [59] in suggesting that fecal calprotectin alone was not reliable for predicting relapse during any given time. Following this, whilst some studies have shown a tendency for serum calprotectin to be higher in IBD patients when compared to other groups [61, 62], very few have been able to show its feasibility for predicting relapse as an independent biomarker [63].

Thus far, the majority of fecal calprotectin research has been aimed at predicting relapse based on clinical markers rather than endoscopic or histological criteria, though a few have sought the latter. In a study designed to evaluate post-surgical recurrence in patients operated on for CD, Orlando

et al. [64] found that a fecal calprotectin cutoff value of 200 mg/L measured at 3 post-operative months gave the best combination of specificity (75 %) and sensitivity (63 %) in predicting endoscopic relapse 12 months after the initial surgical intervention, though the statistical significance of these results was not stated. Interestingly, though the use of an endoscopic definition of relapse in some otherwise asymptomatic patients as opposed to a clinical definition cannot be overstated, the authors [64] did have similar results to some larger studies done previously [37]. Another study looked at a similar cohort of patients, specifically focusing on endoscopic relapse post surgical intervention for a CD patient cohort 12 months after their surgery, and obtained largely disparate results to the former study. Lasson et al. [65] followed 30 patients from the time of their ileocaecal resection until 12 months post-operation and found that not only was there little difference in the mean fecal calprotectin levels of those with endoscopic recurrence (189 μg/g) and those with endoscopic remission (227 μg/g), but that the previously proposed cutoff levels of 200 and 250 μg/g [66, 67] had no predictive value between the endoscopic recurrence and endoscopic remission groups, though these results were not statistically significant. Two recent studies showed that the measurement of fecal calprotectin concentrations is a promising and useful tool for monitoring CD patients after surgery [68, 69]. Several other studies have also looked at the role of fecal calprotectin and CRP for monitoring and predicting postoperative recurrence of IBD (reviewed in [58]).

Lactoferrin

Lactoferrin is a multifunctional iron-binding glycoprotein that constitutes the major component of polymorphonuclear neutrophil granules and is secreted by most mucosal surfaces [70, 71]. The presence of inflammation in the intestinal lumen triggers polymorphonucelar neutrophils to infiltrate the mucosal surfaces, which in turn causes a proportional increase in lactoferrin concentrations in the feces [72, 73]. Given its stability in feces, its ability to be unaffected by multiple freeze–thaw cycles, and the relatively small decline in fecal levels after 2 days at room temperature [74, 75], lactoferrin has been extensively studied as a potential marker of direct intestinal inflammation, with variable outcomes [76, 77].

Since its identification as a potentially useful fecal marker for intestinal inflammation, lactoferrin has been studied multiple times in comparison to more well-known markers (Table 3). Many studies have looked at the correlations of fecal lactoferrin level with certain clinical indices of active disease [78–80], but in one of the first studies comparing the calprotectin and lactoferrin for their predictive capabilities, Gisbert et al. [47] followed a mixed cohort of 163 IBD patients for up to 12 months to assess for clinical relapse as defined by the Truelove modified index [81]. Whilst the sensitivity and specificity of fecal calprotectin using a cutoff value of >150 μg/g to identify the relapse population yielded values of 69 % for both, fecal lactoferrin seemingly showed similar performance, with an overall sensitivity and specificity of 62 and 65 %, respectively, when looking at the IBD cohort as a whole. Interestingly, the authors identified that if one were only to use predictions of relapse for the first 3-month period rather than the whole 12 months, fecal lactoferrin had an overall sensitivity of 100 % with a specificity of 62 %, marking it as a potentially very useful screening tool for early relapse. Following on from this, Yamamoto et al. [79] looked at a smaller cohort of CD patients for 12 months post ileocolonic resection. Similar to the previous studies, their findings supported the notion that both fecal calprotectin and fecal lactoferrin had potential roles in predicting disease recurrence; using a fecal calprotectin cutoff level of 170 μg/g yielded a sensitivity of 83 % and specificity of 93 %, whilst a fecal lactoferrin cutoff value of 140 μg/g gave a sensitivity of 67 % and a specificity of 71 % at 12 months. The authors identified a few limitations of their study, namely the relatively small sample size, and the fact that prophylactic medications had been given to the patients post-operatively on an individual basis to decrease the likelihood of recurrence, and they recommended that future studies aiming to validate the predictive value of fecal biomarkers should be done in patient cohorts with stable medications. Following their own advice, a year later the same group published a study looking at the predictive value of both fecal calprotectin and fecal lactoferrin in UC patients who were in stable remission for >3 months on a stable mesalamine maintenance therapy [82]. After following this cohort for a similar 12-month period, Yamamoto et al. [82] found that using similar cutoff values to their previous study yielded similar results, with a fecal lactoferrin sensitivity and specificity of 67 and 68 %, respectively. However, only fecal calprotectin was found under multivariate analysis to be a predictor of relapse ($P = 0.002$); lactoferrin was not found to be significantly associated with clinical relapse of the disease.

Only a few studies evaluating the predictive value of fecal lactoferrin have been performed in the pediatric population, with mixed outcomes. Turner et al. [83] used a relatively large cohort of 101 pediatric UC patients undergoing intravenous corticosteroid treatments, taking stool samples during their third day of therapy and measuring four prominent fecal biomarkers head-to-head. Using the need for either second-line treatment or colectomy as the definition of failed corticosteroid therapy, Turner et al. [83] found that whilst fecal lactoferrin levels

Table 3 Studies investigating other fecal markers for the prediction of relapse in inflammatory bowel disease

Study	Patient population	Subjects (*n*)	Marker	Cutoff level	Sensitivity/ specificity (%)	PPV/ NPV (%)	Treatments allowed whilst in remission	Relapse definition
Casellas et al. [106]	Adult UC	54	DNA	63,900 copies/μg	77/96	NA	No restrictions	UCAI ≥7
Yamamoto et al. [82]	Adult UC	80	Calprotectin Lactoferrin	CP: 170 μg/g LF: 140 μg/g	CP: 76/76 LF: 67/68	NA	All patients given mesalamine	Worsening of stool frequency and/or rectal bleeding with endoscopic features suggestive of moderate to severe inflammatory changes
Yamamoto et al. [79]	Adult CD	20	Calprotectin Lactoferrin	CP: 170 μg/g LF: 140 μg/g	CP: 83/93 LF: 67/71	83/93 50/83	No restrictions	CDAI >150 with increase of ≥70 points
Walker et al. [84]	Pediatric IBD (≤ 21 years)	CD: 79 UC: 62	Lactoferrin	NA	NA	NA	No restrictions	Clinical deterioration significant enough to warrant a change in medical management
Däbritz et al. [85]	Adult/pediatric IBD	CD: 61 UC: 120	S100A12	IBD: 0.43 mg/kg CD: 0.35 mg/kg UC: 0.43 mg/kg	IBD: 70/83 CD: 67/100 UC: 73/79	NA	No restrictions	CDAI >250 or CDAI >150 with increase of ≥70 points; PCDAI >20 or PCDAI >10 with increase of ≥5 points; UCAI >6 or UCAI >4 with increase of ≥3 points; PUCAI >40 or PUCAI >10 with increase of ≥5 points
Saitoh et al. [92]	Adult IBD	CD: 37 UC: 42	Eosinophil proteins	NA	NA	NA	No restrictions	CDAI >150
Boirivant et al. [90]	Adult CD	11	α-1 Antitrypsin	62 mL/day	NA	100/100	No restrictions	Presence of narrowing of the preanastamotic loop with ulcerations, nodularity, and/or cobblestone appearance
Biancone et al. [91]	Adult CD	26	α-1 Antitrypsin	40 mL/day 120 mL/day	100/15 75/85	19/100 50/94	No restrictions	CDAI >150

IBD inflammatory bowel diseases, *CD* Crohn's disease, *UC* ulcerative colitis, *PPV* positive predictive value, *NPV* negative predictive value, *(P)CDAI* (pediatric) Crohn's disease activity index, *(P)UCAI* (pediatric) ulcerative colitis activity index, *CP* calprotectin, *LF* lactoferrin, *NA* not available

were greatly elevated in the study cohort as a whole, their predictive value in detecting responders and non-responders was limited (median lactoferrin levels were 209 μg/g in responders and 225 μg/g in non-responders, $P = 0.92$). Though in an admittedly different cohort of patients, Walker et al. [84] aimed to identify whether fecal lactoferrin had any prognostic or predictive value in pediatric patients with IBD. Looking at a group of 148 pediatric patients with both UC ($n = 62$), CD ($n = 79$) and IBS ($n = 7$) and comparing them to a healthy control group ($n = 22$), the authors found that not only did fecal lactoferrin correlate comparatively to ESR when aiming to detect clinically active disease ($P < 0.001$), but it also showed a statistically significant trend of being higher in patients who experienced a clinical flare-up of their disease within 2 months (845 μg/mL) compared with those who remained in clinical remission (190 μg/mL, $P = 0.003$).

S100A12

Released under conditions of cell stress, S100A12 (calgranulin C) is a member of the S100 calcium-binding protein family that was recently reported to be a novel biomarker for inflammation based on fecal and serum studies of IBD [85]. More specifically, S100A12 is released almost exclusively by neutrophils following pattern recognition receptor (PRR) activation [77], and it amplifies inflammation through its capacity to act as a ligand on monocyte Toll-like receptor 4 [86].

While the relationship between S100A12 and IBD disease activity is well described elsewhere [30, 87, 88], there have been few studies to date that have investigated the potential for S100A12 to be used as a biomarker for the prediction of relapse in IBD. A 2010 study on the predictive capabilities of a variety of biomarkers by Turner et al. [83] showed S100A12 as having a poor ability to predict steroid refractoriness in their cohort of pediatric patients with severe UC ($P = 0.11$). These findings were certainly less admirable than those for calprotectin, which showed a much stronger predictive capability. However, there has since been further investigation into S100A12 and its role as a biomarker for predicting IBD.

A recent cohort study by Däbritz et al. [85] showed promising results and indicated, for the first time, the potential for fecal S100A12 to predict relapse in UC and CD (Table 3). Däbritz et al. [85] monitored 147 adults and 34 children with CD ($n = 61$) or UC ($n = 120$) who were deemed to be in remission as defined by the (pediatric) CDAI and the (pediatric) ulcerative colitis activity index ([P]UCAI). In the IBD patients who fell back into an active disease process ($n = 62$), fecal S100A12 levels were significantly elevated at inclusion ($P < 0.0001$). In spite of this significant finding relating to fecal levels, the ability of serum S100A12 to predict relapse in both diseases was shown to be poor. However, serum S100A12 levels were still significantly elevated in patients with active CD or UC as compared to those in remission ($P < 0.005$). Däbritz et al. [85] also describe the observation of rising fecal S100A12 levels in relapse patients from below the predictive cutoff value (0.43 mg/kg) 9 months before relapse (median 0.1 mg/kg) to an increased median level of 0.5 mg/kg ($P < 0.001$) 6 months before relapse, and a further increase to 0.9 mg/kg 3 months before relapse ($P < 0.0001$). This further evidence of the predictive capabilities of fecal S100A12 aligned with the median peak relapse values of 5.5 mg/kg ($P < 0.0001$). ROC curve analysis of fecal S100A12 provided sensitivity and specificity values of 70 and 83 % for IBD as a whole. When focusing on CD, sensitivity and specificity levels were shown to be 67 and 100 %, respectively, and they were 73 and 79 %, respectively, for UC. In the same study, S100A12 was shown by Däbritz et al. [85] to have slightly higher specificity and lower sensitivity values than fecal calprotectin for CD and UC. Similar findings were noted upon comparison of the sensitivities and specificities of fecal calprotectin levels reviewed above [36, 37, 40, 45, 46, 64, 85] with those of fecal S100A12 reported by Däbritz et al. [85]. Thus, the future use of S100A12 as non-invasive investigation for predicting IBD relapse shows promise.

Fecal α1-antitrypsin

Lactoferrin, along with the S100 and acute-phase proteins, has been the most intensively investigated biomarker for the predication of relapse in IBD to date. However, there have been a few investigations of other proteins and cytokines too, one of which is fecal α1-antitrypsin (α_1-AT) (Table 3). Fecal α_1-AT levels are known to reflect the degree of protein loss in CD patients and are a sensitive marker of CD activity [89]. Yet there have been limited studies focused on the ability of fecal α_1-AT to predict CD relapse. To begin investigating this, Boirivant et al. [90] followed just 11 resected CD patients over 12 months. After measuring fecal α_1-AT every 3 months, patients who experienced recurrence at the end of the study period ($n = 5$) had increased fecal α_1-AT levels at 6 months post-resection, with the concentrations recorded at this point and at 12 months post-resection higher than those observed in the patients who did not relapse ($P < 0.01$). This relationship between a rising fecal α_1-AT level and CD relapse was studied again in 2003 by Biancone et al. [91], who investigated a slightly larger cohort of 26 patients in remission of ileal CD as defined by CDAI. After taking a baseline fecal α_1-AT level and following patients in a

short-term study of 3 weeks and a long-term study of 3 months, the authors were able to show that median fecal α_1-AT at baseline was significantly higher in the patients who returned to active CD within the next 6 months ($P = 0.03$). The sensitivity and specificity of fecal α_1-AT were 75 and 85 %, respectively, yet the PPV for relapse in the next 6 months was only 50 %, with a NPV of 94 %. This indicates that fecal α_1-AT has some ability to predict disease relapse within 6 months, but has a better NPV.

Eosinophil-derived proteins

Given that eosinophils involved in inflammatory disease are known to produce cytotoxic proteins capable of causing the mucosal damage seen in IBD, Saitoh et al. [92] investigated patients with UC ($n = 42$) and CD ($n = 37$) to assess the value of eosinophil-derived proteins in predicting disease (Table 3). Fecal levels of eosinophil cationic protein (ECP) and eosinophil protein X (EPX) were the study's main focus, with EPX shown to be more stable within the stool sample for 48 h after collection than ECP. Whilst neither ECP nor EPX showed any value in predicting UC relapse, there was a significant difference in the levels of both ECP and EPX between CD patients who did not relapse and those who did within the following 3 months ($P < 0.001$). Critically, Saitoh et al. [92] compared the performance of ECP and EPX to that of lactoferrin, fecal α_1-AT, and fecal hemoglobin. The close correlation between concentrations of these markers and EPX, coupled with its stability, showed that EPX may be useful in predicting a relapse of CD.

Granulocyte macrophage colony-stimulating factor

Having only recently come under scrutiny for its potential role in both immune and inflammatory reactions within the human gastrointestinal tract, granulocyte macrophage colony-stimulating factor (GM-CSF) has been the focal point of few studies to date [93]. A hematopoietic growth factor that promotes activation and maturation of myeloid cell derivatives, GM-CSF has been shown to have dose-dependent proinflammatory effects during the host's response to infection or injury within a wide range of tissues, including lung, intestinal lamina propria, and skin [94]. Although it is known that an increased level of GM-CSF leads to a proinflammatory response, the detailed role of GM-CSF in the intestine of humans is, to date, relatively unknown. However, the results of a clinical trial showing that GM-CSF treatment for IBD led to decreased disease activity in a subgroup of patients with CD [95] have

Table 4 Studies investigating serum surrogate markers for the prediction of relapses in inflammatory bowel disease

Study	Patient population	Subjects (n)	Marker	Cutoff level	Sensitivity/specificity (%)	PPV/NPV (%)	Treatments allowed whilst in remission	Relapse definition
Däbritz et al. [97]	Adult/paediatric IBD	CD: 61 UC: 120	GM-CSF Ab	CD: 1.7 µg/mL UC: 0.5 µg/mL	CD: 88/95 UC: 62/68	NA	No restrictions	ΔCDAI ≥70 points in 2 weeks, ΔPCDAI ≥5 points in 2 weeks, ΔUCAI ≥3 points in 2 weeks, or ΔPUCAI ≥5 points in 2 weeks
Amre et al. [98]	Pediatric CD	139	ASCA pANCA	NA	NA	NA	No restrictions	First clinical event (complication or related surgical intervention)
Lakatos et al. [99]	Adult CD	91	LBP sCD14	22,650 ng/mL 1,395 ng/mL	55/84 62/72	78/65 69/66	No restrictions	CDAI >150 or ΔCDAI >100 with change in medical therapy
Louis et al. [101]	Adult CD	29	IL-2	95 pM/L	NA	NA	No restrictions	CDAI >150 or ΔCDAI >100
Louis et al. [102]	Adult CD	36	IL-6	20 pg/mL	NA	NA	No restrictions	CDAI >150 or ΔCDAI >100
Braegger et al. [107]	Pediatric IBD	CD: 26 UC: 10	TNF-α	NA	NA	NA	No restrictions	NA

IBD inflammatory bowel diseases, *CD* Crohn's disease, *UC* ulcerative colitis, *(P)CDAI* (pediatric) Crohn's disease activity index, *(P)UCAI* (pediatric) ulcerative colitis activity index, *GM-CSF Ab* granulocyte macrophage colony-stimulating factor autoantibodies, *LBP* lipopolysaccharide-binding protein, *sCD14* soluble CD14, *IL* interleukin, *ASCA* anti-*Saccharomyces cerevisiae* antibodies, *pANCA* perinuclear antineutrophil cytoplasmic antibodies, *TNF* tumor necrosis factor, *NA* not available

generated interest in the role of endogenous neutralizing serum GM-CSF autoantibodies (Ab) as a potential biomarker for the prediction of relapse in IBD (Table 4), particularly given the inverse relationship between GM-CSF Ab levels and GM-CSF bioactivity [96].

With only one study investigating such a role for serum GM-CSF Ab so far, further investigation is needed to reaffirm the interesting findings of Däbritz et al. [97]. Time course analysis of a broad cohort with CD ($n = 61$) and UC ($n = 120$) patients showed an obvious increase in the titer of serum GM-CSF Ab 6 months before relapse ($P < 0.0001$), with an initial elevation noted as early as 9 months before the relapse had occurred ($P < 0.01$). Critically, peak values were observed during relapse before a statistically significant return to baseline values 9 months later. Importantly, when considering the potential role of GM-CSF Ab as predictors of relapse, the patients who experienced a CD relapse had higher GM-CSF Ab levels at inclusion than those who remained in remission. Through ROC curve analysis, at 1.72 μg/mL the sensitivity and specificity of GM-CSF Ab for predicting a CD relapse within 6 months before a relapse were found to be 88 and 95 %, respectively—far superior to the predictive capabilities of calprotectin seen in the same study (84 and 50 %, respectively). Whilst these results of Däbritz et al. [97] clearly show a significant association between GM-CSF Ab and CD relapse, the same level of significance was not seen when considering their results on UC. The sensitivity and specificity for UC relapse prediction within 6 months before a relapse of the disease was far less impressive at 62 and 68 %, respectively. What this does show, however, is that there might be a place for GM-CSF Ab to be used as a predictor of CD alongside other biomarkers that are more appropriate for the prediction of UC relapse. From the perspective of a potential clinical application in the future, it was also shown that a baseline level of GM-CSF Ab >1.7 μg/mL was significantly associated with a CD relapse within 18 months.

Antibodies

In 2006, Amre et al. [98] carried out a retrospective cohort study on 139 pediatric patients in order to evaluate the role of serum anti-*Saccharomyces cerevisiae* antibodies (ASCA) and pANCA as predictive markers (Table 4). Assayed close to diagnosis following trial inclusion in the mid-1990s, ASCA-IgG and -IgA along with pANCA status were used to identify a hazard ratio (HR) for complications. Using Cox proportional hazards models, any ASCA positivity suggested a 2.33 HR for a complication at any time during the disease ($P < 0.05$). ASCA-IgA positivity showed a slightly higher risk factor than ASCA-IgG (2.84 and 2.38, respectively, $P < 0.05$). Amre et al. [98] also showed that those who were positive for either ASCA or had a higher titer of ASCA-IgA were more likely to have a complication earlier than those who did not. The authors also described a negative association between pANCA positivity and risk of recurrent complications. Whilst these results show a significant link between ASCA and prediction of CD complications, the number of complications was quite low, with 22 % of the 139 patients experiencing complications during follow-up, and no sensitivity, specificity, NPV, or PPV values were reported. Coupled with the need for a study of adult subjects, a larger cohort needs to be followed to attain more knowledge around the predictive value of ASCA and pANCA.

Lipopolysaccharide-binding protein and soluble CD14

Other novel markers investigated are those focused on by Lakatos et al. [99] in their study of a mixed cohort of 214 CD patients (Table 4). Both lipopolysaccharide-binding protein (LBP) and soluble CD14 (sCD14) play key roles in the innate immune response against the lipopolysaccharide of gram-negative bacteria [100], and the reports of changes in LBP and sCD14 levels in animal models of IBD led to the authors' interest in their utility as predictive markers. During a 1-year follow-up period, 23 % of the 91 remission patients relapsed, and cutoff levels of LBP and sCD14 were used (>22,650 and >1,395 ng/mL, respectively) to calculate odds ratios for identifying relapse. Individually, LBP showed a higher odds ratio (6.5) than sCD14 did (4.3). However, the sensitivity (55 % LBP, 62 % sCD14), specificity (84 % LBP, 72 % sCD14), along with the NPV (65 % LBP, 66 % sCD14), and PPV (78 % LBP, 69 % sCD14) were moderate for both and are by no means significant enough to consider these markers promising candidates for future clinical use at this stage.

Cytokines

Finally, various cytokines and soluble receptor levels have also been investigated for future utility as predictive markers for IBD relapse (Table 4). In 1995, Louis et al. [101] assessed soluble IL-2 receptor (sIL2-R) levels in CD relapse prediction, given its correlation with disease activity. Through a 12-month longitudinal follow-up of 29 patients with inactive CD, it was discovered that a high soluble IL-2 level was predictive of relapse when levels were elevated beyond 95 pM/liter ($P = 0.008$), with the relative risk associated with such an increased level calculated as 9.9. However, Louis et al. [101] explain that whilst their result showed correlations with CDAI and other parameters of inflammation such as ESR and CRP,

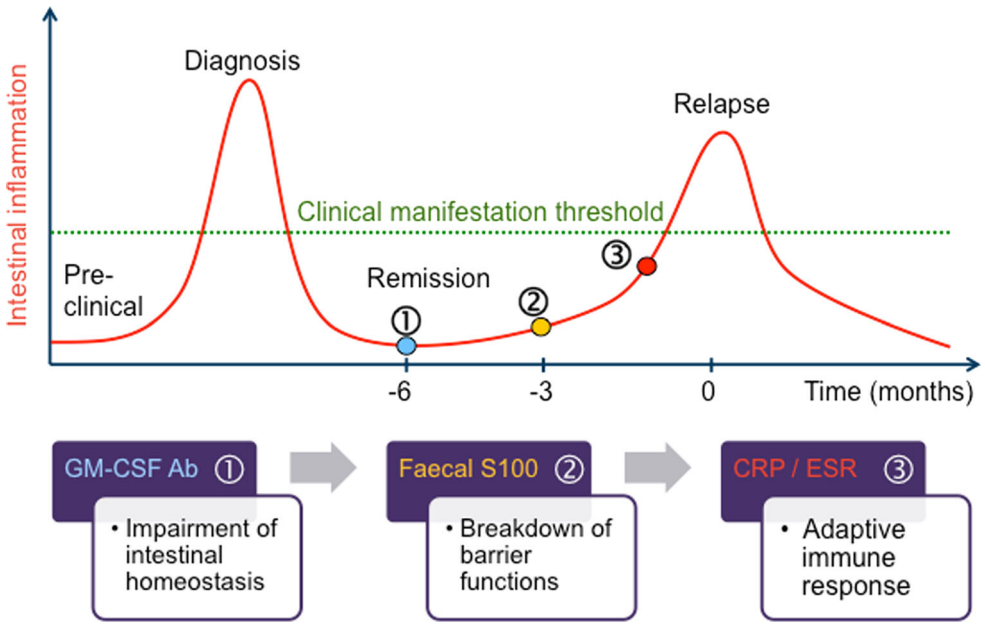

Fig. 1 Biomarkers in the prediction of relapse in inflammatory bowel disease. An outline of disease activity, from preclinical symptoms through to a relapse in disease as indicated by a clinical manifestation threshold. At various stages of remission, the reviewed biomarkers are shown to have different predictive validities, correlating with their increased activity as inflammation in the gastrointestinal tract increases. Granulocyte macrophage colony-stimulating factor (GM-CSF) autoantibody (Ab) levels can predict relapse 6 months prior (*blue dot*); fecal S100 proteins (*yellow dot*) are capable of predicting relapse within 3 months; and C-reactive protein (CRP)/erythrocyte sedimentation rate (ESR) (*red dot*) is only able to predict relapse weeks before it is seen clinically

other investigations have been less consistent. As such, sIL2-R was shown to be complementary to the levels of inflammatory markers in this study, and showed some predicative ability for a relapse of CD within 12 months, but it requires a lot more investigation given that this was the first time that such a finding was reported.

Following on from this 2 years later, Louis et al. [102] investigated the value of IL-6, TNF-α, and soluble TNF receptors as predictive biomarkers for CD relapse. The authors concluded that IL-6 had the greatest predictive value for a relapse within 1 year if serum levels were elevated beyond 20 pg/mL ($P < 0.001$). Only 10 % of patients who had an IL-6 level above 20 pg/mL remained in remission 12 months after their inclusion in the study. While TNF-α itself did not show any predictive value, the two soluble TNF-α receptors p55 and p75 showed some ability to predict relapse in the coming 12 months when their values were above 2 and 4.3 ng/mL, respectively ($P = 0.004$ and $P = 0.04$, respectively). However, similar to their earlier studies, no sensitivity, specificity, NPV, or PPV values were described.

Conclusions

Biomarkers, both serological and fecal, have the potential to form the mainstay of the monitoring of IBD patients whilst they are in remission. There is a tendency to believe that the fecal component of this potential assay is more specific to clinical and subclinical activity in patients suffering from bowel disorders, which is supported by the current body of evidence when compared to other serological acute-phase reactants. Fecal calprotectin has attracted the lion's share of the attention thus far, partly because of initial promising studies that showed a real potential for this marker to be predictive of future relapses. Whilst it has proven to be very effective in its own right, it has not yet shown the high predictive value required for a casual screening test, and has therefore been challenged by new markers. Initial investigations into other fecal biomarkers such as lactoferrin, eosinophil-derived proteins, and α1-AT, as well as serological biomarkers such as the acute-phase reactants, cytokines, ESR, CRP, and other antibodies, have all been compared to fecal calprotectin with variable levels of success. More recently, however, the emergence of new biomarkers such as S100A12 and GM-CSF Ab have added a greater depth of knowledge to the field, and are offering alternate avenues for research and additional predictive tools for assessing the potential for relapse (Fig. 1). Ultimately, for now, no single biomarker—whether it be fecal or serological—has shown the consistency through study to be considered a gold standard; nor has any come close, to be fair. However, enough have shown promise and potential, and given the possibility of creating matrices of predictive tools using this broad range

of biomarkers, there is still room for more investigations into these areas. These biomarkers have the potential to become cornerstones of predictive models for relapse monitoring in IBD, and may yet form the standard of care of our practice one day soon.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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