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What is the role of C-reactive protein and fecal calprotectin in evaluating Crohn's disease activity?

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ABSTRACT

Historically, the evaluation of patients with Crohn's disease (CD) has centered on use of subjective symptom-based assessment. However, patients with CD experience a broad spectrum of non-specific symptoms that may not directly correlate with objective measures of inflammation. Endoscopy has been the gold standard for evaluating the burden and severity of mucosal disease. However, use of ileocolonoscopy for disease monitoring in long-term follow-up is limited by considerations of cost, resource utilization, and invasiveness. As treatment goals in CD have shifted towards 'treat-to-target' paradigms that emphasize tight control of inflammation, it has become increasingly evident that sensitive, accurate, and reliable measures of disease activity are required. The use of non-invasive serum and fecal biomarkers such as C-reactive protein (CRP) and fecal calprotectin (FC) has been evaluated in patients with CD for categorizing disease activity, predicting treatment response, identifying patients at risk for disease relapse, and as a potential therapeutic target. In this review, we summarize the interpretation of CRP and FC in patients with CD within specific clinical contexts and according to assay performance characteristics.

Introduction

Patients with Crohn's disease (CD) experience a wide range of gastrointestinal symptoms, including diarrhea, abdominal pain, rectal bleeding, nausea, vomiting, and weight loss [1]. However, these symptoms are non-specific and consequently, there is a poor correlation between symptomatic and objective measures of disease activity [2,3]. A subset of CD patients have persistent intestinal inflammation that results in silent disease progression despite being asymptomatic [4] while conversely, many patients in endoscopic remission fail to enter clinical remission [5]. Unsurprisingly, targeting symptom resolution alone has largely failed to change the natural history of CD since asymptomatic patients with subclinical inflammation remain untreated or treatment is substantially delayed [6]. Therefore, a "treat-to-target" paradigm that emphasizes early initiation of effective medical therapy targeting validated clinical and objective endpoints has been endorsed. In the Selecting Therapeutic Targets in Inflammatory Bowel Disease (STRIDE) recommendations, resolution of abdominal pain, normalization of stool habit, and absence of ulceration on ileocolonoscopy or cross-sectional imaging in patients with disease beyond the reach of endoscopy are recommended as targets [7].

Although endoscopy has been the mainstay of objective disease evaluation in CD, the feasibility of using ileocolonoscopy repeatedly in long-term follow-up is limited by availability, its invasive nature, patient tolerability, and cost. Therefore, there is substantial interest in using non-invasive biomarkers for monitoring CD activity. C-reactive protein (CRP) and fecal calprotectin (FC) are the most widely used biomarkers for disease evaluation. CRP is a pentameric

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Abbreviations

ACCENT	A Randomized, Double-blind, Placebo-controlled Trial of Anti-TNFα Chimeric Monoclonal Antibody Infliximab in the Long-term Treatment of Patients With Moderately to Severely Active Crohn's Disease
CALM	An Open-Label, Multicenter, Efficacy and Safety Study to Evaluate Two Treatment Algorithms in Subjects With Moderate to Severe Crohn's Disease
CD	Crohn's disease
CDEIS	Crohn's Disease Endoscopic Index of Severity
CHARM	Crohn's Trial of the Fully Human Antibody Adalimumab for Remission Maintenance
CI	confidence interval
CRP	C-reactive protein
DIS	dose intensification strategy
FC	fecal calprotectin
HBI	Harvey Bradshaw Index
HR	hazard ratio
IBD	inflammatory bowel disease
IL	interleukin
MRE	magnetic resonance enterography
NPV	negative predictive value
OR	odds ratio
POCER	Postoperative Crohn's Endoscopic Recurrence
RCT	randomized controlled trial
ROC	receiver operator curve
SES-CD	Simple Endoscopic Score for Crohn's Disease
STRIDE	Selecting Therapeutic Targets in Inflammatory Bowel Disease
TAILORIX	A Randomized Controlled Trial Investigating Tailored Treatment With Infliximab for Active Luminal Crohn's Disease
TDM	therapeutic drug monitoring
TL	trough level
TNF	tumor necrosis factor
UC	ulcerative colitis

protein that is synthesized constitutively in hepatocytes [8]. In CD, CRP is also produced by mesenteric adipocytes [9] and high concentrations of CRP mRNA expression have been shown in the mesenteric fat of patients with CD. CRP synthesis is primarily regulated by interleukin (IL)-6 and serum concentrations increases rapidly within hours of inciting inflammatory stimuli [10]. However, serum CRP concentrations are non-specific, and elevations can be observed with immune-mediated diseases, obesity, trauma, cardiovascular events, infections, and neoplasia [11]. In contrast, fecal biomarkers may have a theoretical benefit of directly reflecting mucosal processes: FC is a calcium- and zinc-binding neutrophil cytosolic protein with anti-bacterial and anti-fungal properties [12]. Cellular degranulation in response to intestinal inflammation results in FC release into the stool, correlating with 111-indium-radiolabeled granulocyte migration [13]. Although FC is correlated with luminal inflammation in patients with either CD or ulcerative colitis (UC), elevations in FC can also be observed in patients with infectious colitis, colorectal cancer, diverticulitis, and those using non-steroidal anti-inflammatories or proton pump inhibitors [14].

Both CRP and FC are increasingly used in clinical practice as adjunctive tools to assess CD activity. The accurate interpretation of CRP and FC depends on the clinical context in which the tests are performed, the performance characteristics of the assay and specified test cut-offs, and the pre-test probability of disease activity. Results should also be interpreted in the context of potential inter- and intra-individual and -assay variability. In this review, we summarize the evidence for using CRP and FC for disease monitoring in CD, assessing response to medical therapy, predicting CD relapse after medically- or surgically-induced remission, and as a potential treatment target.

Evaluating disease activity using biomarkers

For biomarkers to be useful for disease monitoring in patients with CD, they must first be well correlated with objective endoscopic, radiographic, and histologic measures of inflammation. The correlation between CRP and FC with CD activity indices has been evaluated using a wide range of methodologies [15–22]. The reported correlation between these biomarkers and other measures of CD activity is variable and at times, conflicting. For example, CRP appears to be only modestly correlated with CD-related symptoms and although Jones et al. found a significant correlation between CRP and the Simple Endoscopic Score for CD (SES-CD) >7 with an area under the receiver operator curve (ROC) of 0.79, other authors have found poor discrimination using an a SES-CD ≥4 or a Crohn's Disease Endoscopic Index of Severity (CDEIS) score ≥6 as definitions of active disease [16,20,21]. Differences in endpoint definition or assay performance likely explain some of these discordant findings.

The diagnostic accuracy of CRP and FC for active inflammatory disease defined by an endoscopic gold standard in symptomatic patients was evaluated in a meta-analysis by Mosli et al. [23] The authors identified only 19 studies evaluating biomarker measurement performance against an endoscopic gold standard despite screening over 2500 records. In the pooled analysis, an elevated CRP was only 49% sensitive [95% CI: 34%, 64%] for endoscopically active CD although specificity was high (0.92 [95% CI: 0.72, 0.98]). In comparison, an elevated FC was 87% sensitive [95% CI: 82%, 91%] and 67% specific [95% CI: 58%, 75%] for endoscopically active CD. Interpretation of these measurement characteristics is dependent on the FC cut-offs used. In this meta-analysis, thresholds adopted by the original study authors ranged from 57 to 280 μg/g. Similar sensitivity (0.81 [95% CI: 0.77, 0.84] and specificity (0.81 [95% CI: 0.76, 0.86]) values were reported in a meta-analysis by Lin et al. that evaluated FC in 727 patients with CD. In their analyses, an area under the ROC of 0.88 [95% CI: 0.83, 0.93] was estimated [24].

Whether FCP more accurately identifies patients with endoscopically active disease than CRP in a population of patients who are known to mount a CRP response is unknown. In a study performed by Schoepfer et al., FC showed better rates of correct classification of endoscopic disease activity defined as inactive (SES-CD 0–3), mild (SES-CD 4–10), moderate (SES-CD 11–19), and severe CD (SES-CD ≥20) compared to CRP [20]. When a low FC cut-off is chosen (e.g. 50–70 μg/g), FC has a higher sensitivity, specificity, and negative predictive value (NPV) for endoscopically active disease (SES-CD ≥4) compared to CRP ≥5 mg/L [20]. Furthermore, FC has been correlated with both magnetic resonance enterography (MRE) measures of inflammation on including mural edema and ulceration [25], and histologic features such as basal plasmacytosis, lamina propria/epithelial polymorphonuclear cellular infiltration, and granulomas [26,27]. In a pediatric cohort of 151 CD patients, FC also accurately classified patients with mucosal healing (SES-CD <3), transmural healing (MRE visual analogue score <20 mm), and deep healing (combined endoscopic and radiographic remission)

[28]. Despite these advantages, patient compliance with repeated stool collection is a disadvantage of FC compared to CRP. Patients perceive stool testing as more difficult to perform and less acceptable than venipuncture [29] although compliance with the former may be improved by point-of-care, home testing [30].

Some authors have raised concerns about poor measurement performance of FC in patients with isolated ileal or small bowel CD [31]. A lower sensitivity of FC for detecting small bowel CD has been hypothesized to be secondary to more proximal disease location and potentially smaller affected mucosal surface area. In support of this concept Gecse et al. showed that patients with large ulcerations (>5 mm) in the ileum had significantly lower FC concentrations compared to patients with ileocolonic or colonic disease (297 vs. 1523 μg/g, $p < 0.0001$) [32]. However, this liability issue may be overcome by adapting lower cut-offs for FC interpretation. In cohort studies comparing FC with either balloon-assisted endoscopy or video capsule endoscopy, FC remained sensitive for detecting active small bowel inflammation when lower thresholds were used. However, this modification might compromise test specificity. Kopylov et al. pooled data from seven studies and 463 patients with small bowel CD: a FC concentration >50 μg/g had 83% sensitivity and 53% specificity for active CD defined by capsule endoscopy [33]. Bressler et al. have proposed FC thresholds for clinical practice. Patients with a FC concentration <50–100 μg/g are likely to have quiescent disease whereas patients with FC concentration >250 μg/g likely have active inflammation [14]. Notwithstanding these recommendations, based upon the totality of published data the optimal cut-off for FC for defining endoscopic disease activity in CD remains uncertain. The use of combined testing has been proposed as a means of increasing biomarker accuracy. Patients with an indeterminate FC concentration 100–250 μg/g are in the uncertain range and in these cases, combining FC with CRP or clinical symptoms may improve the sensitivity for detection of endoscopic inflammation [34]. Likewise, a prediction model comprised of FC, fecal immunochemical test for stool hemoglobin, and symptoms defined endoscopic healing in CD patients with 86% specificity [35]. Improving the diagnostic accuracy of non-invasive testing though the use of multiple biomarker panels is a promising approach that requires validation in prospective cohort studies.

Predicting treatment response using biomarkers

Changes in biomarker concentrations have been evaluated as a potential predictor of response to medical therapy. This concept is especially pertinent during induction therapy when biomarker changes may occur earlier than resolution of mucosal ulcers by endoscopy.

Conflicting results have been reported for the use of CRP to predict treatment response. Nevertheless, CRP has been used to stratify patients based on inflammatory burden in clinical trials. Support for this concept comes from the observation that an inverse correlation was observed between baseline CRP concentration and response to placebo in trials of certolizumab and ustekinumab [36,37]. Interestingly in some studies, patients with a high baseline CRP concentration have been shown to have higher response rates to active therapy [38,39]. For example, in a post-hoc analysis of the CHARM (Crohn's Trial of the Fully Human Antibody Adalimumab for Remission Maintenance) randomized controlled trial (RCT), patients with a baseline CRP concentration ≥10 mg/L were more than twice as likely to achieve week 56 clinical remission with adalimumab therapy compared to patients with a low baseline CRP concentration (47% vs. 23%, $p < 0.05$) [40]. However, the opposite effect has been reported in observational studies that show an inverse correlation between high baseline CRP concentration and clinical response [41–43]. These conflicting results may be reconciled by differences in study design. In clinical trials, high CRP at baseline selects for patients with more active disease who are likely to respond to immunosuppression as compared to those with functional symptoms who may respond equally well to placebo [44]. In contrast, patients in observational studies with a high CRP concentration at baseline may have greater inflammatory burden that is less likely to respond to medical therapy. Additionally, it should be recognized that CRP response may be specific to the treatment mechanism, independent of any beneficial effect on endoscopically-defined inflammation. For example, tumor necrosis factor (TNF) directly up-regulates IL6 production that, in turn, result in a rapid reduction of CRP. Accordingly, treatment with TNF antagonists rapidly decrease serum concentrations of TNF whereas similar decreases may not be observed for agents that do not directly block TNF such as vedolizumab [45,46].

Dynamic changes in CRP and FC with medical therapy may be a better predictor of treatment response than baseline concentrations. In the ACCENT I trial (A Randomized, Double-blind, Placebo-controlled Trial of Anti-TNFα Chimeric Monoclonal Antibody Infliximab in the Long-term Treatment of Patients With Moderately to Severely Active Crohn's Disease, a ≥60% reduction in CRP concentration at week 14 amongst patients with an elevated baseline CRP levels predicted sustained response through week 54 (odds ratio OR 7.3 [95% CI: 1.4, 36.7]) and patients with CRP normalization <5 mg/L by week 14 were more than twice as likely to maintain response (OR 2.20 [95% CI: 1.26, 3.85]) [38,47]. Similarly, normalization of FC ≤ 100 μg/g after induction therapy has been shown to be highly associated with the likelihood of clinical remission (84% vs. 38%, $p < 0.0001$) [48] at one year whereas failure to suppress FC after infliximab induction was associated with treatment discontinuation [49]. Sipponen et al. demonstrated that median FC levels drop significantly if endoscopic response is achieved (73 μg/g vs. 1282 μg/g, $p = 0.005$), irrespective of the class of medical therapy [50]. However, given that these observations are primarily retrospective and uncontrolled; confirmatory prospective studies are required.

Biomarkers are increasingly used as a secondary endpoint in CD clinical trials. The use of biomarkers in combination with clinical symptoms may help increase differentiation between active comparator and placebo groups in distinction to symptom-based outcomes alone. For example, Sands et al. compared the efficacy and safety of the anti-IL23p19 monoclonal antibody MEDI2070 to placebo in a phase II RCT [51]. At week 12, there was a statistically insignificant difference in clinical response rates (defined by a 100-point decrease in the Crohn's Disease Activity Index (CDAI) score) between patients treated with MEDI2070 and placebo (37.3% vs. 28.3%, $p = 0.29$). However, when a composite endpoint comprised of clinical response and a ≥50% reduction in either CRP or FC was evaluated, a significant treatment effect was observed in favor of MEDI2070 (37.3% vs. 8.3%, $p < 0.001$). Combining biomarker and symptom-based endpoints may therefore improve trial efficiency by increasing the separation between active treatment and placebo response rates compared to CDAI-defined outcome criterion alone.

Predicting disease relapse based upon biomarkers

An area of growing interest is the use of biomarkers for risk stratification of asymptomatic CD patients. The validity of this concept, which relies on the accurate detection of prognostically relevant pre-clinical inflammation prior to the onset of symptoms, is supported by several observations. First, serum or fecal biomarker changes may reflect the pathological processes occurring in the intestinal wall. For example, high FC concentrations have been associated with transmural diffusion restriction abnormalities on MRE in asymptomatic patients [52]. Second, an association has

been shown between elevated biomarker concentrations in asymptomatic patients with subsequent risk of histologic, endoscopic, and symptomatic relapse [53]. Therefore, it is postulated that measuring biomarker concentrations during maintenance therapy could provide a window in which to initiate additional investigations or proactively intensify treatment with the expectation that such actions would reduce the risk of disease-related morbidity.

Serial measurement of CRP may be used to predict adverse outcomes. Bhattacharya et al. followed 185 CD patients who were in clinical remission, defined as a Harvey Bradshaw Index (HBI) $\leq$4 and short Inflammatory Bowel Disease (IBD) Questionnaire score $\geq$50, for a minimum of four years [54]. Patients with a CRP concentration >7.4 mg/L developed new structural damage as measured by the Lemann index at a higher rate (65% vs. 36%, $p < 0.0001$) and were more likely to require hospitalization (median 1 vs. 0 admissions, $p = 0.005$) over the duration of follow-up compared to patients with a persistently low CRP concentration. Likewise, in a prospective registry of 339 CD patients in clinical remission defined by CDAI <150 for >6 months, Oh et al. showed that a CRP concentration >6.0 mg/L was associated with an increased risk of both intestinal resection (hazard ratio HR 1.73 [95% CI: 1.00, 2.97]) and hospitalization (HR 1.79 [95% CI: 1.25, 2.57]) [55], suggesting that CRP elevations even in patients in clinical remission portend a negative prognosis.

Mao et al. evaluated the operating characteristics of FC for predicting disease relapse in a meta-analysis of six studies including 354 patients with quiescent CD [56]. An elevated FC (>50-340 μg/g, depending on the original study author definition) had a sensitivity and specificity for relapse of 0.75 [95% CI: 0.64, 0.84] and 0.71 [95% CI: 0.64, 0.76], respectively. When FC determination was combined with CRP or therapeutic drug monitoring (TDM), the PPV for identifying patients at high risk for disease relapse increased. Roblin et al. demonstrated that the combination of subtherapeutic infliximab trough levels (<2 μg/mL) and an elevated FC concentration (>250 μg/g) accurately identified 87% of CD patients who relapsed over a mean follow-up duration of 20.4 months [57]. Serial measurement of biomarkers may also improve test performance: in a prospective cohort of 71 CD patients in clinical remission receiving TNF antagonists who were followed with every 4-month FC testing, the overall probability of disease relapse was 31%. However if on consecutive testing the FC concentration was >300 μg/g, the rate increased to 86% [58]. In contrast, a meta-analysis of six studies that included 552 IBD patients demonstrated that serially normal FC tests was associated with a reduction in risk of relapse compared to those with elevated values of only 6–20% [59]. Multiple issues remain to be addressed including definition of the optimal measurement interval; measuring more frequently than every three months may be limited by practical considerations, particularly patient acceptability, whereas measuring less frequently may be ineffective [60].

Similar considerations exist for CD patients in surgically-induced remission, however in this circumstance the use of biomarkers may be even more cogent because symptom-based measures do not correlate with endoscopic disease recurrence [61]. Existing studies indicate while that CRP is neither sensitive nor specific for detection of endoscopic disease recurrence in CD patients, FC performs somewhat better. In the POCER (Postoperative Crohn's Endoscopic Recurrence) study, patients with a Rutgeerts score $\geq$ i2 had higher post-operative FC compared to patients who remained in endoscopic remission (275 vs. 72 μg/g, $p < 0.001$) [62]. A FC concentration >100 μg/g was highly sensitive for endoscopic recurrence (89%), however the specificity was poor (58%). The authors suggested that FC testing could reduce surveillance colonoscopies by 47% post-operatively at the cost of a 4% false negative rate. In comparison, the performance characteristics of FC for predicting endoscopic relapse in a large multicenter prospective trial of postoperative mercaptopurine compared to placebo were less impressive [63]. The pooled AUC of FC for identifying of endoscopic recurrence was only 0.70 [95% CI: 0.63–0.77] and although FC predicted endoscopic recurrence with high sensitivity (84%) at a cut-off of 50 μg/g, specificity was only 44%. Additionally, when the cut-off was increased to 100 μg/g, the NPV of FC for endoscopic recurrence was only 76% (as compared to the >90% estimated in POCER). A meta-analysis of 10 prospective studies that evaluated 613 patients with CD in post-operative remission confirmed that while an elevated FC concentration is highly sensitivity for endoscopic recurrence (0.82 [95% CI: 0.73, 0.89]), the specificity is poor (0.61 [95% CI: 0.51, 0.71]) [64]. Thus, taken collectively, the evidence does not support the use of a single biomarker in isolation for appraising the risk of postoperative recurrence; however, multiple tests in combination may be able to optimize the sensitivity and specificity for disease relapse.

Treating to biomarker targets

In recent years, the “treat to target” concept, which intensifies therapy until a clearly defined therapeutic goal is achieved, has become accepted for management of IBD. Although the STRIDE Consensus guidelines do not recommend biomarker-defined treatment targets, there are supportive data emerging that these tests can play a useful role in treatment algorithms. The CALM (An Open-Label, Multicenter, Efficacy and Safety Study to Evaluate Two Treatment Algorithms in Subjects With Moderate to Severe Crohn's Disease) trial compared a biomarker-based tight control algorithm to a conventional clinical symptom-based strategy in patients with moderate-to-severe CD [65]. Patients with clinical, biomarker, and endoscopic activity at baseline (defined by a CDAI 150–450 depending on concurrent prednisone dosing, CRP $\geq$5 mg/L or FC $\geq$ 250 μg/g, and CDEIS $\geq$6 and sum of CDEIS sub-scores $\geq$6 in one or more segments with ulcers) were randomized to either treatment intensification using biomarkers or clinical criteria. Patients in the biomarker-guided group received treatment intensification based upon both symptoms (CDAI $\geq$150 or prednisone use) or an elevated CRP $\geq$5 mg/L or FC $\geq$ 250 μg/g. Conversely, treatment in the clinical management group was only intensified if a patient met symptom-based criteria (CDAI decrease <70 or CDAI >200 prior to randomization, CDAI decrease <100 or CDAI >200 or prednisone use post-randomization). Treatment was either intensified or de-escalated based on pre-specified algorithms at weeks 12, 24, and 36 with sample procurement one week prior to evaluation. The algorithm for treatment intensification was escalation from no treatment, to adalimumab every other week, to adalimumab weekly, to combination therapy with both adalimumab and azathioprine.

Compared to the symptom-based management group, a higher proportion of patients in the biomarker-guided treatment group achieved mucosal healing (CDEIS <4 without deep ulcers) (46% vs. 30%, $p = 0.01$). Moreover, a higher proportion of patients in the biomarker-guided treatment group also achieved deep remission (CDAI <150, CDEIS <4 and no deep ulcers, absence of draining fistula, discontinuation of corticosteroids $\geq$8 weeks), biological remission (FC < 250 μg/g, CRP <5 mg/L, and CDEIS <4), corticosteroid-free remission, and clinical remission (CDAI <150). Treatment intensification in the biomarker-guided group was prompted by an increased FC concentration in 62%, 56% and 45% of patients at weeks 11, 23 and 35, respectively. In contrast, CRP prompted treatment escalation in 46% of patients at weeks 11 and

23 and 45% at week 35.

These results suggest that tailoring treatment to biomarker-based targets is feasible and associated with improved patient-outcomes in comparison to clinical management alone, however the study had some relevant limitations. First, a recommendation for a single biomarker target cannot be identified because treatment escalation in CALM was triggered using both CRP and FC. Second, CDAI targets were different in the clinical compared to the tight-control groups. Third, patients enrolled in CALM had very early disease, and were naïve to both TNF antagonists and immunosuppressants. The effect of treating to biomarker targets in CD patients with established CD is unclear. Fourth, the effect of treating with subsequent entry biologics, combination biologic therapy, or novel oral small molecule therapies is unclear. Finally, it is uncertain whether treating to biomarker endpoints is superior to targeting endoscopic endpoints (e.g. absence of ulcers), which is currently being evaluated in the REACT2 (Enhanced Algorithm for Crohn's Treatment Incorporating Early Combination Therapy) study (NCT01698307).

Targeting biomarker endpoints in combination with TDM has been evaluated in the TAILORIX (A Randomized Controlled Trial Investigating Tailored Treatment With Infliximab for Active Luminal Crohn's Disease) study [66]. In this RCT, biologic-naïve CD patients treated with infliximab were randomized to one of three maintenance algorithms based on symptoms, biomarkers, and TDM compared to conventional symptom-based management alone. Two dose intensification strategies (DIS) were evaluated (DIS1 and DIS2). In all patients the following criteria, in hierarchal order prompted dose escalation: (1) CDAI >220 with a CRP >5 mg/L and/or FC > 250 μg/g; (2) CDAI 150–220 for two consecutive weeks with an elevated CRP and/or FC; (3) infliximab serum trough level (TL) < 1 mg/mL; (4) infliximab TL 1–3 mg/mL and 5) infliximab TL 3–10 mg/mL with a drop by >50% compared with the week 14 infliximab concentration. An additional infliximab infusion at 4-week intervals was administered if the TL was <1 mg/mL in both DIS groups. However, in the DIS1 group, infliximab was escalated by 2.5 mg/kg increments up to twice, whereas a 5 mg/kg increase was given once in the DIS2 group. The control group received infliximab dose increases of 5–10 mg/kg based on clinical symptoms (CDAI >220 or a CDAI 150–220 in the two prior visits) without biomarker use or TDM. Corticosteroid-free clinical remission without ulcers, need for surgery, or the development of fistulas between weeks 22–54 was evaluated as the primary outcome.

Dose escalation occurred in 44% (20/45), 62% (23/37), and 40% (16/40) of patients in the DIS1, DIS2, and control groups, respectively. In the DIS1 and DIS2 groups, 5 and 7 patients received dose-escalated based on TDM; 9 and 12 patients dose escalated based on a combination of clinical symptoms and biomarker elevations, respectively. In contrast, 15/16 patients in the control group received dose escalation based exclusively on their CDAI score. No difference in the proportion of patients achieving the primary outcome was observed (33%, 27%, and 40% of patients in the DIS1, DIS2, and the control group, respectively; $p = 0.50$). Furthermore, no significant differences were observed in the proportion of patients who achieved secondary endpoints of absence of ulcers, endoscopic remission or endoscopic improvement at both week 12 and 54. Although outcomes did not differ between the DIS and control groups, several issues limit the interpretation of the TAILORIX study. Importantly, fewer than half of patients in the DIS1 (47%) and DIS2 (46%) groups had sustained therapeutic infliximab TL > 3 mg/mL between weeks 12 and 54. Second, dose escalation algorithms in TAILORIX incorporated TDM, biomarkers, and symptoms: the independent effects of each of these components is difficult to evaluate.

Conclusions

CRP and FC are now widely used in the management of patients with CD. However, their interpretation is dependent on the specific clinical circumstance in which the tests are applied. Both tests have strengths and limitations. In patients with established CD, CRP and FC may be used to non-invasively evaluate inflammatory disease activity and are moderately correlated with endoscopic, radiographic, and histologic measures of inflammation. Both tests have limited specificity that constrains their use as an alternative to endoscopy for making major treatment decisions. Rather, CRP and FC provide adjunctive information for predicting treatment response and disease relapse and may serve as a trigger for endoscopic assessment. Promising findings from the CALM trial suggest that biomarkers may be a surrogate treatment target; however, additional data from large scale studies are required to determine the efficacy and cost-effectiveness of this approach relative to one based upon endoscopic outcomes.

Finally, it is important to be aware that clinical interpretation of CRP and FC results is dependent on how the operating properties of the test modify the pre-test to post-test probability of active disease. For example, when the pre-test probability of active CD is uncertain (i.e. 50%), a normal FC of 35 μg/g results in a 16% post-test probability of disease; conversely, a high FC of 350 μg/g would result in a post-test probability of endoscopic inflammation of 73% [23]. Are these post-test probabilities sufficiently acceptable to both the clinician and the patient to make a treatment change? It is plausible that neither party would be able to accept a 16% chance of missing active inflammation or escalate therapy with a 27% chance of overtreating endoscopic remission. In this example, the FC result, either positive or negative, does not defer the need for additional investigations.

Clearly, the literature evaluating the operating characteristics of CRP and FC is highly diverse and heterogeneous. Comparing results across different study designs, patient populations, and test assays is impossible. Nevertheless, the evidence overall suggests that CRP and FC can be helpful biomarkers for identifying intestinal inflammation in patients with CD, though predominantly in a supportive rather than primary role for evaluating disease activity, monitoring response to treatment, and predicting disease relapse. Although currently limited by modest sensitivity and specificity, CRP and FC may be incorporated with other serum, genetic, metabolic, or proteomic biomarker panels to improve accuracy for disease assessment and prognostication in the future.

Practice points

- CRP and FC are useful adjunctive tests for assessing inflammatory CD disease activity
- Biomarker reductions after initiation of medical therapy may be associated with treatment response
- Serial measurement of biomarkers may help identify patients at high risk for disease relapse or post-operative recurrence

Research agenda

- Comparative long-term outcomes between treating to endoscopic targets versus biomarker targets need to be clarified
- Future studies should determine the accuracy of using multiple biomarker panels in combination for evaluating disease activity

Conflicts of interest

- C.M., R.B., and C.E.P. have no conflicts of interest to declare.
- R.K. has received consulting/speaker fees from AbbVie, Encycle, Janssen, Merck, Pendopharm, Pfizer, Roche, Robarts Clinical Trials, Shire, and Takeda Canada.
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- V.J. has received has received consulting fees from AbbVie, Eli Lilly, GlaxoSmithKline, Arena pharmaceuticals, Genetech, Pendopharm, Sandoz, Merck, Takeda, Janssen, Robarts Clinical Trials, Topivert, Celltrion; speaker's fees from Takeda, Janssen, Shire, Ferring, Abbvie, Pfizer.

Author contributions

CM, RB, RK, CEP, BGF, and VJ contributed to manuscript drafting and critical revision for important intellectual content.

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