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Expert recommendations regarding the use of fecal calprotectin in daily clinical practice: statements of a taskforce process from the Hellenic Group for the Study of Inflammatory Bowel Disease (EOMIFNE)

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Abstract

Fecal calprotectin (FC) has become an indispensable tool in everyday clinical practice for the diagnosis and management of inflammatory bowel disease (IBD). Nonetheless, specific and clear recommendations for its use are scarce. On behalf of the Hellenic Group for the Study of IBD (EOMIFNE), a group of experts have formulated 18 statements with the aim to provide evidence for the best use of FC and offer practical guidance in common clinical scenarios. Statements address issues for the proper application of FC measurement in the diagnostic workup of patients with gastrointestinal symptoms, and as a screening tool for the management of established IBD, including pediatric patients. Thus, the present manuscript aims to propose a standardized approach for the optimal use of FC in patient care, also taking into account non-medical concerns.

Key words Fecal calprotectin, inflammatory bowel disease, EOMIFNE, guidelines

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Introduction

Calprotectin constitutes the major cytosolic protein of neutrophils, and is released from those cells in response to pathogen-associated molecular patterns or danger-associated molecular patterns. Its concentration is an indicator of tissue infiltration by neutrophils, which signifies the presence of active inflammation. Accordingly, fecal calprotectin (FC) is secreted by infiltrating neutrophils that are present in the intestinal tissue during inflammatory processes, including inflammatory bowel disease (IBD); thus, measurement of FC has emerged in recent years as an accurate and valuable noninvasive biomarker for gastrointestinal inflammation [1,2]. Those characteristics have led to the inclusion of FC in numerous national and international guidelines, and the proposal for its use: a) as a screening tool for the diagnosis of IBD; and b) as a monitoring instrument in patients with established IBD [3-5].

Despite the ample existing literature that supports the applicability and clinical relevance of measurement of FC in patients with IBD, the test is not currently reimbursed in Greece. This may in part be due to the fact that the optimal use of FC in everyday practice has not yet been clearly defined, including its applicability in specific clinical scenarios. To address these unmet needs, the Hellenic Group for the Study of IBD (EOMIFNE) organized a taskforce with the goal of

formulating expert recommendations that aim: a) to clarify "gray areas" and dictate clinical decisions about the optimal use of FC for the management of Greek patients; b) to serve as a practical guide for gastroenterologists, but also other specialists (primary care physicians, rheumatologists, internists); and c) facilitate regulatory agencies in decision-making regarding the implementation of FC measurements in the reimbursement plan of the National Health system in Greece.

Methodology

A taskforce was organized consisting of two senior IBD experts as chairpersons (GB and MT), and several active EOMIFNE members who were assigned specific predefined subtopics of major interest related to the use of FC. The core members of this taskforce considered that strict application of the GRADE methodology was impractical, and proposed the use of a hybrid approach, which consisted of a systematic literature review in the context of a modified GRADE system, expert discussion, and voting to form consensus recommendations. Details regarding the way the level of evidence and strength of recommendations were derived are summarized in Table 1.

For each topic of interest, a thorough literature search was conducted for relevant publications in the PubMed, EMBASE and MEDLINE databases. The selection of published papers for further analysis was based on specific inclusion and exclusion criteria. In particular, only articles written in the English language were included. In addition, priority was given to papers with higher levels of evidence, such as randomized controlled trials (RCTs) and meta-analyses (MA). Finally, more recent publications were also given higher priority.

This evidence-based process resulted in the development of a draft containing several conditional recommendations for the use of FC. This first draft of statements was circulated for additional comments among all EOMIFNE members. A final online meeting was then scheduled, with the participation of an expert panel of 30 adult and pediatric gastroenterologists. During this meeting, additional refinement of the statements took place, followed by voting for each statement. Final approval was granted only to those statements that achieved an 80% rate of agreement among the expert panelists.

Results

Technical aspects of FC measurement

We recommend the use of fresh stool samples for FC measurements. If this is not feasible, samples can be stored at 4°C for up to 1 week until final analysis. (Strong recommendation, Consensus 100%)

Studies consistently report significant FC degradation in samples stored at room temperature (25°C), which can begin as early as within the first 24 h [6,7]. Consequent decreases in FC concentration correlate to the duration of storage prior to analysis [6-10] and can reach up to 50% after 1 week [7]. To mitigate this reduction, samples should be stored at 4°C until analysis, as data support a clear superiority of refrigeration [6,7,10], with reported preservation of stable calprotectin values for up to 1 week post-collection [6]. Calprotectin extraction using buffers has been shown to enhance the maintenance of stable values after 1 week when stored at 4°C [5,6], but there are equivocal results for their storage at room temperature. However, the longevity of FC regarding its degradation in the commercially available extraction buffers is highly dependent on the assay manufacturer's specifications. Hence, all procedures should be conducted in accordance with the respective instructions [7]. Deep freezing (<-20°C)

Table 1 Details on how levels of evidence and strength of recommendations were derived using a hybrid approach, incorporating both the modified GRADE system and voting outcomes

Level of evidence	High	Evidence derived from meta-analyses or systematic reviews of randomized controlled trials (RCTs), or multiple high-quality RCTs with consistent results directly applicable to the clinical question
	Moderate	Evidence derived from a single RCT, well-designed prospective cohort studies, or meta-analyses with moderate heterogeneity or indirectness
	Low	Evidence derived from retrospective cohort studies, case–control studies, small prospective studies, or *post hoc* analyses
Consensus process	Voting panel	A multidisciplinary panel of 30 adult and pediatric gastroenterologists with expertise in IBD participated in the final voting process
	Consensus threshold	Agreement by ≥80% of panelists was required for statement approval
Strength of recommendation – hybrid approach	Strong recommendation	Strong recommendations were supported by current data with high or moderate levels of evidence, or by low levels of evidence supplemented by clear clinical experience (as reflected by >90% agreement among the voting panel)
	Conditional recommendation/ suggestion	The taskforce was not sufficiently confident to provide a strong recommendation, based on current data; however, a conditional recommendation/suggestion can be provided as a clinical tool, in the context of current common clinical experience

of samples is superior to refrigeration [9], and it is preferred for sample preservation longer than 1 week, since stable calprotectin values can be maintained for 6-12 weeks.

We recommend that a single measurement of FC at a specific time point is adequate for clinical decision-making, as day-to-day, intra-day, and intra-assay variability do not significantly impact the result. (Strong recommendation, Consensus 96%)

Significant intra-day variability in FC values has been observed, although not uniformly. In several studies, diurnal fluctuation of FC was reported to be significant [8,11-13] and was attributed to various factors. Lasson *et al* reported statistically significant elevations in FC levels during evening bowel movements and in looser stools, and a trend for higher values in bloody stools [8], although the correlation between elevated FC values and evening bowel movements was not confirmed in other studies [13,14]. It is noteworthy that this intra-day variation is more profound in cohorts of patients with active IBD [12,13], even though it does not result in clinically meaningful variations. In contrast to diurnal variation, intra-assay and day-to-day variability appear to be negligible. In particular, when FC levels from the same bowel movement were measured at 2 distinct time points, to calculate intra-assay variability, the resulting values demonstrated a strong intraclass correlation of 0.79 [10] and 0.91 [15]. Day-to-day variability was reported in only 1 cohort of 63 patients, with low clinical relevance (9% for the 200 μg/g and 5% for the 50 μg/g threshold [16]. In other studies, day-to-day variability was either absent [15,17] or not statistically significant [14]. It should be noted that variation may be particularly pronounced at higher FC values, yet instances of deviations concerning patient positivity are infrequent. Therefore, it is reasonable to conclude that a single measurement of FC is adequate in clinical practice.

We recommend that quantitative FC testing, when available, should be preferred over qualitative assays. Enzyme-linked immunosorbent assays (ELISA) and automated ELISA platforms in certified laboratories offer the most accurate measurements. Point-of-care (POC) and at-home testing devices are considered reliable alternatives. (Strong recommendation, Consensus 97%)

Currently, various immunobiochemistry methods that target proteinic epitopes in the stools are available for the measurement of FC concentration. Pretreatment of stool samples is required before measuring FC, to allow for the extraction of the protein in the appropriate buffer [18]. The standard method is the weighing method, which includes weighing of a certain amount of stool to achieve the required concentration in a buffer; however, this is time-consuming and impractical for a busy laboratory to adopt [18]. To address these issues, multiple specialized, commercially available extraction/measurement kits have been developed [18]. In a relevant study, the fluorescence-immunoassay EliA Stool Extraction Kit (EliA SEK, Thermo Fisher, Uppsala, Sweden) and the Calex® Cap (Calex, Bühlmann Laboratories AG, Schönenbuch, Switzerland) extraction device, which uses particle-enhanced turbidimetric immunoassay (PETIA) with BühlmannfCAL® Turbo assay (Bühlmann Laboratories AG, Schönenbuch, Switzerland), were compared against the weighing extraction method with favorable results [19]. To minimize the effect of liquid stool consistency in the evaluation of FC concentration, an additional step of dilution of a standard volume of liquid stool in the respective buffer volume, and centrifugation to exclude the sediment of the analysis can be considered [20,21]. It is also recommended to perform 4 extractions from 4 different locations of the stool specimen to ensure a representative FC concentration estimation on further analysis, as extraction from a single stool location leads to high heterogeneity for the same sample [19].

FC can be estimated using both qualitative (reporting a positive/negative result), and quantitative (reporting the exact concentration of FC in stools) methods [19]. Quantitative measurements are recommended in patients with IBD, given the high clinical relevance of accurate calculations of the precise concentration of FC [22]. The ELISA method, is considered the gold standard for FC measurement [23]. Multiple automated ELISA-based platforms have been developed (fluoro-enzyme immunoassays, chemiluminescence immunoassays, and particle-enhanced turbidimetric immunoassays), that need less laboratory manipulation time and can even be carried out individually for each sample. These assays are reliable, and correlate significantly with the standard ELISA methods, as they provide equivalent results [8,9,13,24-34].

Recently, POC devices have been developed, with strong correlation to ELISA platforms and good accuracy profile, thereby providing reliable results and enabling on-site quantitative or semi-quantitative (within a range of values) FC level estimation [23,35-43] within 30 min [37]. Another useful tool for monitoring patients with IBD are at-home, patient-processed testing devices, which also utilize immunochromatography and produce semi-quantitative results [44]. The result is uploaded to a corresponding application of the specific kit via a smartphone camera and sent to the treating physician. These assays demonstrate significant correlation to ELISA-based assays and POC devices [44-51]. It should be noted that proper patient training regarding the use of these medical devices is crucial for producing reliable results.

We recommend that the same assay should be used for sequential measurements of FC during follow up, given the high variability among different methods. (Strong recommendation, Consensus 100%)

Inter-assay differences in FC measurements values are commonly observed, and are characteristic of nearly all assay comparisons. In a study that compared 4 different assays—EliA calprotectin CN (Thermo Fisher Scientific), EliA calprotectin CN2 (Thermo Fisher Scientific), Liaison DiaSorin calprotectin (DiaSorin, S.p.A.), and Bühlmann fCAL turbo (Bühlmann Laboratories AG)—a high correlation was reported, although

significant proportional bias was observed, especially at high FC values [26]. Similar findings for the comparison between FC-specific immunoassays have also been reported by other groups [32-34,40].

Consequently, although variability exists among the available assays, there is no conclusive evidence supporting the superiority of any particular assay. Thus, we do not advocate for the use of any specific assay. On the other hand, given the aforementioned inter-assay variability it is highly recommended that serial measurements of the same patient should preferably be made using the same assay.

FC as a screening tool for IBD (Table 2)

We recommend against the use of FC testing as a screening test in asymptomatic individuals. (Strong recommendation, strong level of evidence, Consensus 100%)

FC testing is a valuable tool for assessing intestinal inflammation, particularly in differentiating IBD from functional disorders such as irritable bowel syndrome. However, its application is not universal, and certain clinical scenarios warrant caution or alternative approaches [52]. Routine FC testing in asymptomatic individuals is not recommended. Elevated FC levels in the absence of symptoms may not correlate with significant pathology, and can lead to unnecessary investigations. Studies have shown that, while FC is a sensitive marker for inflammation, its specificity decreases in asymptomatic populations, leading to potential overdiagnosis of an inflammatory syndrome [5,53]. In patients with acute diarrhea (<4 weeks), especially when infectious etiologies are suspected, FC testing is not advised as a primary diagnostic tool. Acute infections can increase FC levels, but this does not distinguish between self-limiting infections and chronic inflammatory conditions. The Infectious Diseases Society of America suggests that FC/lactoferrin detection should not be used to establish the cause of acute infectious diarrhea, and there are insufficient data on the value of FC measurement in this context [54].

For patients over 50 years of age, or those presenting with alarm symptoms (e.g., unexplained weight loss, rectal bleeding, anemia), relying solely on FC testing is inadequate. While FC can be elevated in conditions other than inflammation, such as polyposis or neoplasms, its accuracy in detecting gastrointestinal malignancy is low. Endoscopy remains the gold standard for evaluating mucosal pathology and detecting intestinal pathology in high-risk groups, and should not be replaced by FC testing [54].

Table 2 Summary of recommended cutoff values and corresponding clinical interpretations by scenario

	Patient population	FC cutoff (μg/g)	Clinical interpretation/recommended action	Strength of recommendation
Screening	Adults	<50	IBD unlikely; functional disorder favored	Strong
Chronic GI symptoms (>4 weeks), low malignancy risk		≥50	Further evaluation for IBD warranted	Strong
Monitoring in established IBD (adults)				
Clinical remission	UC/CD	≤150	Low risk of relapse; continue routine monitoring	Strong
Clinical remission with persistently elevated FC	UC/CD	>150	Endoscopic and/or imaging assessment recommended	Strong
Symptomatic patients	UC/CD	≥150	Further objective assessment before treatment escalation	Strong
Post-operative Crohn's disease (6-12 months)	CD	>150	Suggestive of post-operative recurrence, but does not replace endoscopy	Strong
Spondyloarthritis with GI symptoms	SpA	No standardized cut-off	Elevated FC suggests need for further GI evaluation	Conditional
Pregnancy	IBD	Same as non-pregnant adults	FC reliable for monitoring disease activity	Strong
Pediatric patients				
Chronic GI symptoms	Children	≥100	Suggestive of organic disease/IBD	Strong
	≥4 years	No cutoff	Age-related variability; interpret with caution	—
	<4 years			
Pediatric IBD in clinical remission	UC/CD	≤100	Continue monitoring	Strong
Pediatric IBD in remission with persistent elevation	UC/CD	>300	Endoscopic evaluation suggested	Strong

FC, fecal calprotectin; GI, gastrointestinal; IBD, inflammatory bowel disease; UC, ulcerative colitis; CD, Crohn's disease; SpA, spondyloarthritis

We recommend the use of FC testing with a cutoff of 50 μg/g to distinguish between IBD and functional disorders in patients with chronic (>4 weeks) abdominal symptoms suggestive of intestinal inflammation and a low risk for gastrointestinal malignancy. (Strong recommendation, strong level of evidence Consensus 100%)

In patients with chronic (>4 weeks) abdominal symptoms and low risk for malignancy, FC testing can help to distinguish between IBD and functional disorders. Incorporating FC testing into clinical practice for evaluating patients with lower gastrointestinal symptoms can reduce the number of unnecessary referrals for endoscopy, thereby decreasing healthcare costs [1,53-55]. FC concentrations in healthy individuals typically range between 10 and 50 μg/g of stool, though this range can vary, depending on study cohorts and the assays used. The optimal cutoff value for FC testing, above which patients should be referred for colonoscopy, remains a topic of debate [53,55].

Although, the first meta-analysis demonstrated a cut-off level of 100 μg/g as the one with the highest accuracy to detect patients with IBD [56,57], subsequent evidence has supported a lower threshold, above which patients should be referred for further investigations. More specifically, a subsequent meta-analysis by van Rheenen *et al* [58] determined that the optimal cutoff point is 50 μg/g, with FC values below this threshold yielding pooled sensitivity and specificity of 0.93 and 0.96, respectively, making it unlikely that these patients have IBD. Additionally, several other studies have stipulated that threshold, including the British Society of Gastroenterology guidelines, as the likelihood of an IBD diagnosis at that range of values is very low [55,59-61].

Moreover, several essays have determined that higher levels of FC (>100 μg/g), are associated with higher diagnostic accuracy, but lower negative predictive values, resulting in higher rates of false negative results [62-64] leading to delayed diagnosis of IBD [58]. An even lower FC cutoff concentration of 40 μg/g has been shown to result in a very low false-negative rate (≤1%), according to a meta-analysis subsequently performed by Menees *et al* [65]. On the other hand, only 1 recent meta-analysis indicated that the difference in sensitivity and specificity between FC testing cutoffs of 50 μg/g and 100 μg/g was not statistically significant, suggesting that a threshold of 100 μg/g could be adopted as a pre-endoscopy screening tool [66]. In view of these conflicting results, in most cases, a cutoff of 50 μg/g is selected, as it seems to be the most reliable cutoff value.

FC as a monitoring tool in IBD (Table 2)

We recommend measuring FC at least twice annually, as a monitoring tool in patients with IBD in clinical remission. (Strong recommendation, moderate level of evidence, Consensus 100%)

Although no dedicated RCT exists to compare a symptom-alone vs. a biomarker plus symptom-based monitoring strategy in asymptomatic patients with ulcerative colitis (UC), indirect data from observational studies suggest that the latter strategy may offer advantages in the management of these patients. In a meta-analysis of 17 observational studies, including more than 1200 UC patients, performed in the context of the recent American Gastroenterological Association guidelines, it was shown that patients with elevated FC compared to those without had a relative risk of 4.4 (95% confidence interval [CI] 3.48-5.47) to experience disease relapse within a year [3]. The cutoff used by most, but not all, studies was FC>150 μg/g. In another recent meta-analysis of 24 prospective studies that aimed to identify the ideal cutoff for FC to predict relapse in IBD, an FC of 152 μg/g was the threshold shown to perform best. A subgroup analysis of the effect of IBD type (UC or Crohn's disease [CD]) on that outcome showed similar results for both types [67].

Concerning CD, there is a similar lack of dedicated RCTs comparing symptoms alone versus a biomarker in conjunction with symptom monitoring strategy in asymptomatic patients with CD. The landmark CALM study had such a design, but it was conducted in patients with active disease on study entry [68]. Likewise, the more recently published STARDUST trial followed a similar design pattern, with the addition of early endoscopic assessment; nevertheless, the subjects had active symptoms at study initiation [69]. Thus, only indirect data are available to address the optimal use of FC as a biomarker for the management of patients with asymptomatic CD. As mentioned above, in a recent MA of 24 prospective studies that aimed to identify the ideal cutoff for FC to predict relapse in IBD, an FC of 152 μg/g was the threshold with the best performance, while subgroup analysis did not exhibit a significant difference between UC and CD patients [67]. Furthermore, from a collection of 12 cohort studies with close to 1000 CD patients it was reported that, in those with overt elevation of FC (>200 μg/g), there was an almost 5-fold increased risk for disease relapse [4]. However, there is the caveat that there was a high level of heterogeneity between these studies.

Taking into account the aforementioned evidence, it seems reasonable to incorporate FC in the follow up of asymptomatic patients with IBD, with a proposed frequency of at least twice yearly to correspond with the usual schedule of clinical monitoring.

We recommend endoscopic and/or imaging assessment of disease activity, rather than empiric treatment adjustment, in patients with IBD in clinical remission but persistently elevated FC>150 mg/g. (Strong recommendation, moderate level of evidence Consensus 97%)

There is no RCT addressing the question of whether different cutoffs for any biomarker, including FC, should affect therapeutic decisions with regard to treatment modifications and long-term outcomes. Nonetheless, taking into consideration the correlation between FC and important outcomes such as endoscopic healing, we reviewed studies assessing whether there is an optimal cutoff to define endoscopic activity, which is a recognized surrogate marker for worse long-term outcomes, demanding action from treating physicians.

In a retrospective study including 181 UC patients, FC was significantly correlated with both Mayo endoscopy score (MES) and UC endoscopic index of severity (UCEIS), although the association was stronger for the latter. Receiver operating characteristic (ROC) curve analysis indicated an FC cutoff level of 187.0 mg/kg for predicting complete mucosal healing (MH), defined as an MES value of 0 or a UCEIS value of 0. The sensitivity and specificity of a cutoff value of 187.0 mg/kg for complete MH were 0.857 and 0.891, respectively. FC predicts complete mucosal healing and correlates better with the UCEIS than with the MES in patients with UC [70]. Theede *et al* identified an FC cutoff of 192 μg/g for predicting MH by either an MES or UCEIS of 0, with similar sensitivity (75% and 79%, respectively) and specificity (88% and 87%, respectively) [71]. In a recent cross-sectional study from the UK, including UC patients, the ideal thresholds for FC to identify MH, defined as an MES of 0 and a UCEIS <2, were 112 and 148 μg/g, with accuracy of 86.9% and 86.8%, respectively [72].

Regarding CD, there is again a lack of RCTs, but we have results from 11 cohort studies showing that, for the endpoint of endoscopically active disease (defined as an SES-CD score ≥3) or endoscopic remission (SES-CD<3), the optimal sensitivity and specificity for a cutoff of 150±50 mg/g were found to be 81% (95%CI 74-87%) and 72% (95%CI 61-81%), respectively [4]. Moreover, from a recent meta-analysis including 25 studies and over 2500 CD patients, albeit with marked heterogeneity in FC cutoffs (the majority being >200 μg/g) and endoscopic endpoints, FC shows a pooled sensitivity of 81% and specificity of 74% for detecting endoscopic active disease, possibly denoting that FC is a valuable diagnostic tool across different phenotypes of CD [73]. Another recent cross-sectional study of 153 CD patients reported an optimal FC threshold of <92.9 μg/g, with a specificity of 89% and a sensitivity of 77% for detecting endoscopic activity, and proposed the combination of FC with intestinal ultrasound as an even better intervention [74]. In an older MA of 25 prospective observational studies assessing the role of FC in identifying disease activity in IBD patients, the authors found that FC had a pooled sensitivity of 85% and specificity of 75% in suggesting endoscopic activity. In a subgroup analysis studying 3 different cutoff levels (50, up to 100 and >100 μg/g) the authors suggested that the best sensitivity (90.6%) was achieved at 50 μg/g—thus, this is the ideal threshold to be used in clinical practice—whereas the best specificity (78.2%) was found at levels >100 μg/g [75]. The interpretation of this finding is that the higher the cutoff, the lower the sensitivity and the higher the specificity. Thus, we decided, based on additional data from the aforementioned studies, to propose a larger threshold in order to diminish false positive results and subsequently decrease unnecessary colonoscopies.

Nevertheless, given that the reliability of the test in a scenario with low pretest probability—as in the case where there is a discrepancy between symptoms and FC measurement—is lower, repeat measurement of FC values as an alternative in due time is a reasonable decision.

We suggest that treatment adjustment in symptomatic patients with IBD should not rely on symptoms alone, but should also take FC measurements into consideration. (Strong recommendation, moderate level of evidence, Consensus 96%)

We suggest further assessment, rather than empiric treatment adjustment, in symptomatic patients with IBD and FC >150 μg/g. (Strong recommendation, moderate level of evidence, Consensus 88%)

No RCT could be retrieved to compare a symptom-alone versus a biomarker/symptom-based monitoring strategy in symptomatic patients with UC. Nonetheless, indirect data from observational studies indicate that the latter strategy could offer advantages in the management of patients with UC. Symptoms have long been shown to be insufficient to reliably detect endoscopic activity. Indeed, in an individual patient data analysis from 6 registrational studies attempting to assess the relationship of Patient Reported Outcomes (PROs) with endoscopic activity, it was shown that there is a substantial percentage of patients (~10-15%) with Rectal Bleeding Subscore (RBS) 2/3 and Stool Frequency Subscore (SFS) 2/3 who had endoscopic remission (MES 0/1) [76]. In a patient-level analysis of 22 prospective observational cohort studies and 1 post hoc analysis of a phase 3 study, Dulai *et al* attempted to incorporate FC in clinical practice, along with the assessment of PROs. They only assessed predefined FC cutoff levels of ≤50(±10) μg/g or ≥250(±20) μg/g. Indeed, they showed that in the scenario of RBS 2/3 + SFS 2/3, an FC≥250 μg/g greatly diminishes the false positive rate to a percentage of less than 5%, thus potentially avoiding endoscopy [77]. This study shows that incorporating a more objective biomarker in addition to symptoms in the management of patients with UC improves important clinical outcomes.

In CD results are available from the CALM study, as mentioned earlier. This was an open-label RCT recruiting adult patients with moderate to severely active, non-stricturing, non-penetrating CD. Furthermore, these patients were symptomatic (with compounding biochemical and endoscopic results indicative of inflammation) at recruitment, and were also naïve to advanced treatment. After initial treatment with steroids, participants were randomized to a "tight control" group in which treatment escalation was based on biomarkers (including FC>250 mg/g) and/or symptoms, or a "clinical management" group in which treatment escalation was based on symptoms alone. Assessment was carried out per study protocol every 3 months. A total of 244 patients were included, with the "tight control" group showing better results than the "clinical management" group (37% vs. 23%) in achieving all study endpoints by 48 weeks [71]. On the other hand, in the STARDUST trial, timely escalation of ustekinumab treatment in CD patients, based on biomarkers and early endoscopic response, along with clinical symptoms, did not result in significantly better endoscopic outcomes at week 48 than symptom-driven decisions alone [72]. Thus, there seems to be a gap in our knowledge concerning the exact patient phenotype that will benefit the most from additional monitoring.

Currently, there is no consensus regarding the ideal FC cutoff that may accurately indicate the presence of inflammation in symptomatic patients with UC and CD. Indirect data suggest that a threshold of 150 μg/g may indicate the need for endoscopic assessment, and possibly a change in therapy, as it can reliably detect moderate-to-severe inflammation in the presence of symptoms, with an acceptable false positive rate. A higher cutoff may further lower false positive rates, but may inappropriately increase false negative cases. On the other hand, a lower cutoff would increase sensitivity. However, taking into consideration that this is a high pretest probability scenario (symptoms indicative of a flare plus raised FC), the fall in the percentage of false negatives would not be adequate to justify a further decrease in our proposed cutoff. We aimed to propose a cutoff that would provide an acceptable equilibrium between sensitivity, specificity, positive and negative predictive values.

There is also the probability that the combination of FC with novel utilities, such as intestinal ultrasound, will facilitate clinical-decision making for each individual patient.

Use of FC in special populations (Table 2)

We recommend against the use of FC measurement as a substitute for the endoscopic assessment of postoperative recurrence (POR) at 6-12 months after ileocecal resection in patients with CD. (Strong recommendation, low level of evidence, Consensus 100%)

Despite the evolution in IBD treatment, a high proportion of patients with CD (about 50% in the first decade) will still require surgical treatment. After surgery, risk-stratified care management should be implemented. High-risk patients receive treatment to prevent POR, and endoscopic evaluation is recommended 6-12 months postoperatively, aiming to optimize treatment and prevent further disease progression and complications [78].

FC has been tested in the postoperative setting, demonstrating satisfactory accuracy in detecting and/or predicting POR. Several prospective observational studies have shown a positive association between FC concentration and severity of POR (Rutgeerts score), with only rare exceptions [79-82]. Two meta-analyses have evaluated FC's performance in the detection of POR. Qiu *et al*, in 2015, included 10 studies with a total of 613 patients, and reported a pooled sensitivity of 82% (95%CI 73-89%), and a negative likelihood ratio of 0.29 (95%CI 0.197-0.44), although specificity was modest 61% (95%CI 51-71%) [83]. The authors did not propose a definitive FC threshold because of the diverse cutoff values in the included studies. A subsequent meta-analysis in 2018 that included 9 studies and 588 patients found an FC value of >150 μg/g to have the best overall accuracy for predicting POR, with a pooled sensitivity of 70% (95%CI 59-81%), specificity 69% (95%CI 61-77%) and an area under the ROC curve of 0.73 [84].

However, the available evidence is not sufficient to recommend FC measurement as a sole reliable alternative to colonoscopic evaluation 6-12 months postoperatively. In particular, the low sensitivity of FC still suggests the need for endoscopic assessment. This may be attributed to certain limitations in existing studies, including substantial heterogeneity regarding the timing of measurements, POR definition (Rutgeerts Score ≥i2 or ≥i2b), type of assay used, and variability of cutoff values. Further, larger, well designed prospective studies are needed to help overcome these limitations. Currently, FC can be considered in the postoperative setting to assess POR along with other imaging modalities [intestinal ultrasound (IUS), magnetic resonance enterography, small bowel capsule endoscopy] in case endoscopy is not feasible or acceptable, or prior to index postoperative endoscopic evaluation, at 3-6 months (as a single measurement, or even better as serial measurements) to guide the timing of endoscopy [5]. Elevated FC values (typically/commonly >100-150 μg/g) suggest POR and an earlier colonoscopy should be considered, whereas lower values may support delayed or standard colonoscopy timing [80, 84]. Besides incorporating FC monitoring into routine postoperative care, serial measurements thereafter can help track inflammatory activity over time and reduce the need for frequent colonoscopies.

We recommend against the use of FC measurement as a substitute for the endoscopic assessment of inflammatory activity in symptomatic patients with UC who have undergone total proctocolectomy with ileal pouch anal anastomosis (IPAA). (Strong recommendation, low level of evidence, Consensus 100%)

In patients with UC who have a history of IPAA there is emerging evidence in favor of the use of FC in diagnosing pouchitis [85-87]. Although studies have shown a positive correlation of FC with the pouchitis disease activity index, as well as its subscores, several limitations—including small sample sizes, and significant variability and heterogeneity in study design and methodology, as well as in the FC assays used—have not allowed for standardization of its use [88,89].

Pouchoscopy remains the gold standard, not only allowing the distinction between inflammatory and functional pouch disorders (which may both be present, with non-specific overlapping symptoms), but also providing the opportunity to exclude causes of secondary inflammation. FC may be used for the early detection of subclinical inflammation in asymptomatic patients, or in case of symptom onset—either complementary to pouchoscopy (offering values for future comparison), or if an empiric treatment approach is preferred in order to diminish unnecessary antibiotic use [88]. In this case a low cutoff value for ruling out pouchitis (such as FC<50 μg/g) may be used [90].

We suggest the use of FC testing in patients with spondyloarthritis and concomitant gastrointestinal (GI) symptoms as a screening tool for latent IBD. (Conditional recommendation, low level of evidence, Consensus 80%)

Spondyloarthritis (SpA) is a family of inflammatory rheumatic diseases that include ankylosing spondylitis (AS),

psoriatic arthritis (PsA), reactive arthritis, IBD-related arthritis (enteropathic), enthesitis-related juvenile idiopathic arthritis, and undifferentiated types of spondyloarthritis that share a common etiology and immunopathogenesis with IBD [91].

Compared to other non-SpA rheumatic and musculoskeletal disorders, up to two thirds of patients with differentiated SpA have been found to have subclinical GI inflammation [92]. FC is a biomarker that can detect gut inflammation with high sensitivity and specificity in this scenario. In a meta-analysis by Fauny *et al*, 21.2-70.7% of SpA patients were found to have elevated FC levels. Higher FC levels correlated with both the presence of macroscopic (11-80% of patients) and microscopic (41.7-100%) inflammation, but not with GI symptoms [92]. Despite this high frequency of endoscopic/microscopic inflammation, only 6-13% of patients with SpA will eventually develop evident IBD during their disease course [92].

FC measurement could help clinicians to decide who would benefit from a more invasive examination (endoscopy, imaging, capsule endoscopy) and, in case of abnormal findings, a tighter control or a more effective treatment targeting both gut and joint inflammation should be implemented. This management could also aid in reducing unnecessary investigations. However, patients with prominent symptoms suggestive of IBD (e.g., persistent/intermittent abdominal pain or diarrhea >4-6 weeks) should undergo an earlier colonoscopy without the need for a prior elevated FC measurement.

As for optimal cutoff levels, no recommendation can be made because there is insufficient evidence. Values of 266 μg/kg and 132 μg/kg at baseline have been correlated with CD diagnosis (sensitivity 100% and 66.7%, specificity 78.7% and 76.9%, respectively) whereas lower values (85 and 100 μg/kg) were associated with the presence of asymptomatic intestinal inflammation [92]. The same applies to the optimal frequency of testing. However, when interpreting FC values in this population, the possible effect of concomitant use of non-steroidal anti-inflammatory drugs (NSAIDs) or biologics [93-95] should be taken into consideration. NSAIDs might cause mild to moderate reversible FC elevations [96], while anti-tumor necrosis factor alpha agents, effective for both SpA and IBD, may lower FC values and conceal possible coexisting IBD [97]. Caution should also be taken in SpA patients under treatment with anti-interleukin (IL)-17, since it has been associated with GI disorders and even new-onset IBD, although this has not been confirmed as statistically significant on meta-analysis [98]. Clinical screening for IBD and FC testing in the presence of symptoms or risk factors should be applied prior to anti-IL-17 initiation [99].

We recommend the use of FC testing to monitor disease activity during pregnancy in patients with IBD. (Strong recommendation, high level of evidence, Consensus 100%)

No difference has been reported in FC concentrations between pregnant and non-pregnant healthy women. In contrast to other serum biomarkers (hemoglobin, erythrocyte sedimentation rate, C-reactive protein [CRP], albumin, iron), whose values are often affected by the physiological adaption that occurs during pregnancy, FC remains stable without significant fluctuation throughout all trimesters [100,101]. Therefore, it is considered an accurate and reliable noninvasive biomarker for the initial evaluation of chronic GI disorders during pregnancy.

Concerning IBD patients, although pregnancy has occasionally been associated with diminished inflammation [102], close monitoring is mandatory, given the potentially unfavorable impact of disease activity on maternal and fetal outcomes. FC has been proven superior to other biomarkers, since it correlates with disease activity as measured by physician global assessment and clinical disease scores in all trimesters [103]. FC values increase up to 3 months before a clinically evident flare, increasing clinician awareness and leading to subsequent timely management (investigation and treatment modifications) so as to prevent worse outcomes during gestation [104]. Along with symptomatic assessment, the measurement of other serum biomarkers and the emerging use of IUS, FC is a valuable tool for close monitoring of women with IBD during preconception and at each trimester [105]. Thus, endoscopy, despite being a relatively low-risk procedure, may be limited to those patients with an absolute indication for further examination.

FC use in pediatric patients with IBD (Table 2)

A) For children older than 4 years, we suggest using an FC cutoff level of 100 mg/g to distinguish IBD from functional GI disorders (FGID)
B) No recommendation can be made regarding the FC cutoff level in children younger than 4 years old
C) We recommend endoscopy in pediatric patients when there is strong suspicion of IBD, regardless of FC levels (strong recommendation, high level of evidence, Consensus 96%)

It is well recognized that FC values vary with age, and are higher in children than in adults. Numerous studies have examined a broad range of age groups and utilized various statistical methods, identifying a distinction between children under 4 years old and those over 4 years old [106,107,108]. Davidson and Lock, in a large study, evaluated FC levels in 8676 healthy children within 2 different age groups, revealing a median FC of 77 mg/g for children aged 1-3.9 years, 62 mg/g for those aged 4-17.9 years, and 61 mg/g for individuals over 18 years. There were no notable differences observed between the age groups of 4-17.9 years and those over 18 years [106]. In 2 more recent studies, Roca *et al* created valuable nomograms from regression analysis results. In the first study, 174 healthy children aged 0-12 years were categorized into 3 distinct age groups: 0-12 months, 1-4 years and 4-12 years. The cutoff values determined for these 2 groups, based on the lowest 95th percentile for FC in each category, were 910, 286 and 54 mg/g, respectively. Significant variability was observed among individuals below 12 months old [109]. In the second study, which included 212 healthy children aged 4-16 years, it was found that the median concentration of FC for the whole study population, i.e. 18.8 mg/kg, was below the cutoff value

of 50 mg/kg proposed for adults, but approximately 20% of healthy children had concentrations above 50 mg/kg [110]. In the biggest study to date, Kolho *et al* evaluated FC levels in 11,255 healthy children aged 0-18 years. The median level of FC was 51 mg/kg in infants <1 year of age. This was 3-4-fold higher when compared to yearly age groups from 1-10 years (total number of children 5691). Across yearly age groups from 11-18, the median values varied from 11-19 mg/kg (total number of children 5325). Thus, they concluded that FC values in children beyond the first year of life are in general low, and are comparable in children and adolescents [111].

GI symptoms are common in primary care, with FGID being more frequent than organic (OGID). Both types can present with similar symptoms, making diagnosis challenging. Distinguishing between OGID and FGID can be challenging, as their symptoms often lack specificity and tend to overlap. A recent systematic review and meta-analysis by An *et al*, which included 18 studies, found that for distinguishing patients with OGID (including IBD) from those with FGID (16 studies), the estimated sensitivity of FC testing was 81% and the specificity 81% [66]. For distinguishing IBD from FGID (10 studies), sensitivity was 88% and specificity 72%. Assuming a population prevalence for OGID of 1%, the positive predictive value was 4.2% and the negative predictive value 100%. The difference in sensitivity and specificity between FC testing cutoffs of 50 μg/g and 100 μg/g was not statistically significant. The meta-analysis concluded that FC testing can reduce unnecessary colonoscopies and specialist referrals for children with FGID, and should be incorporated into clinical practice for evaluating patients with lower GI symptoms. In another systematic review and meta-analysis, Holtman *et al* concluded that FC, along with CRP and albumin, are more reliable indicators of the need for endoscopy in a child compared to symptoms, while among the former measures FC was more reliable than blood tests [112]. Furthermore, Degraeuwe *et al*, in their meta-analysis of 9 studies and over 800 patients, found that with an FC cutoff level of 50 μg/g there would be 17% false positive and 2% false negative results, concluding that in high-prevalence circumstances, FC can be used as a noninvasive biomarker of pediatric IBD with only a small risk of missing cases, and can help in selecting patients for endoscopic evaluation [113].

We recommend including FC measurement, at least twice annually, for the assessment of pediatric patients with IBD in clinical remission (strong recommendation, high level of evidence, Consensus 100%)

Monitoring FC in IBD patients soon after the initiation of treatment can provide an objective indication of potential clinical progress [52]. Maintaining low levels during treatment is associated with a higher probability of sustaining clinical remission for an additional year [114]. According to Molander *et al*, FC levels below 100 μg/g in IBD patients after the induction phase with infliximab treatment may serve as a positive prognostic indicator for achieving clinical remission [115]. Taking into account the aforementioned evidence, as well as extrapolation from adult data, it seems reasonable to incorporate FC in the follow up of pediatric patients with IBD in clinical remission, with a proposed interval of at least twice yearly.

We suggest endoscopic evaluation in pediatric patients with IBD in clinical remission with persistent FC measurements of >300 μg/g (strong recommendation, low level of evidence, Consensus 92%)

Further studies on FC levels have demonstrated that, in pediatric cases of IBD, the values for predicting disease flares in conservatively treated patients range between 400 and 800 μg/g [114-116]. Notably, FC levels tend to increase prior to any clinical or endoscopic sign of relapse, as observed in a cohort of adult IBD patients who achieved remission confirmed by endoscopy and were monitored using FC levels. The researchers concluded that FC can serve as a useful tool for identifying patients who need more frequent follow up in clinical settings [115]. Thus, in pediatric IBD patients in clinical remission with persistent FC values ≥300 μg/g, endoscopic assessment seems to be a reasonable option. We arbitrarily selected this cutoff, considering that pediatric studies report a flare prediction at higher concentrations (400-800 μg/g) but also recognizing the risk of missing subclinical inflammation at lower values. Thus, this was considered a reasonable compromise threshold, balancing sensitivity and specificity and aiming to reduce unnecessary procedures.

We recommend against the use of FC in children: 1) presenting with an episode of acute gastroenteritis to distinguish between bacterial and viral pathogens; 2) with a suspicion of intestinal polyps; and 3) as a biomarker for the diagnosis or monitoring of celiac disease (strong recommendation, moderate level of evidence, Consensus 100%)

Sykora *et al* indicated that FC can help distinguish between bacterial and viral causes of acute gastroenteritis in children under the age of 3 [117]. Their study demonstrated that when FC is used in conjunction with CRP, the diagnostic accuracy for differentiating between bacterial and viral acute gastroenteritis reached as high as 94%. Similarly, Duman *et al* found that FC levels were significantly elevated in patients with positive results from stool culture, particularly in cases of confirmed bacterial gastroenteritis. In assessing bacterial acute gastroenteritis, they reported that the area under the ROC curve for FC was 0.867, with a sensitivity of 88.9% and specificity of 76.0% at a cutoff of 710 mg/L [118]. Conversely, among children with acute gastroenteritis requiring hospitalization, no significant differences were observed in FC performance between those with a rotavirus and those with *Salmonella* enteritis infections. The higher levels of FC in rotavirus acute gastroenteritis noted in this study can be partially attributed to the study's context, as only hospitalized patients, who are likely to exhibit more severe GI inflammation than those treated in primary care, were included. To conclude, although some studies suggest that FC may differentiate bacterial from viral gastroenteritis

in children, the results are inconsistent, largely derived from hospitalized cohorts, and not validated in primary care or large-scale prospective trials. Furthermore, the significant overlap of FC levels between bacterial and viral infections, as well as the influence of confounding factors (e.g., age, stool consistency, comorbidities), limit its diagnostic accuracy in everyday pediatric practice.

Until recently, there have only been a few sporadic reports and series documenting cases of children with chronic jejunal polyps and elevated FC levels [119,120]. Nevertheless, an increase in FC levels alone cannot differentiate polyps from IBD without colonoscopy. FC levels are markedly higher in patients diagnosed with celiac disease, particularly among those exhibiting elevated serological markers or typical symptoms. However, no relationship has been identified with histological results. Additionally, conflicting findings have emerged regarding the link between FC and anti-tissue transglutaminase levels. Although the average FC values across the studies fluctuated around 100 μg/g, celiac disease patients showed significantly higher results at the time of diagnosis compared to control subjects. This difference diminished within 4 to 12 months after the initiation of a gluten-free diet [121,122]. Thus, the ambivalence of the above-described results concerning the use of FC in these conditions does not allow for a recommendation in favor of using this biomarker.

Concluding remarks

The EOMIFNE taskforce aimed to generate practical and concise statements that will allow for a standardized approach to the incorporation of FC into everyday patient care. We believe that, with the use of a robust hybrid methodology utilizing existing knowledge, along with expert consensus, while also taking account of the specific characteristics of the Greek healthcare system, we have managed to provide herein a clear guiding framework for the optimal use of FC as a useful diagnostic tool, leading to improved patient outcomes.

References

1. Jukic A, Bakiri L, Wagner EF, Tilg H, Adolph TE. Calprotectin: from biomarker to biological function. *Gut* 2021;**70**:1978-1988.
2. Ikhtaire S, Shajib MS, Reinisch W, Khan WI. Fecal calprotectin: its scope and utility in the management of inflammatory bowel disease. *J Gastroenterol* 2016;**51**:434-446.
3. Singh S, Ananthakrishnan AN, Nguyen NH, et al; AGA Clinical Guidelines Committee. Electronic address: clinicalpractice@gastro.org. AGA clinical practice guideline on the role of biomarkers for the management of ulcerative colitis. *Gastroenterology* 2023;**164**:344-372.
4. Ananthakrishnan AN, Adler J, Chachu KA, et al; AGA Clinical Guidelines Committee. Electronic address: clinicalpractice@gastro.org. AGA clinical practice guideline on the role of biomarkers for the management of Crohn's disease. *Gastroenterology* 2023;**165**:1367-1399.
5. Kucharzik T, Taylor S, Allocca M, et al. ECCO-ESGAR-ESP-IBUS guideline on diagnostics and monitoring of patients with inflammatory bowel disease: part 1. *J Crohns Colitis* 2025;**19**:jjaf106.
6. Haisma SM, van Rheenen PF, Wagenmakers L, Muller Kobold A. Calprotectin instability may lead to undertreatment in children with IBD. *Arch Dis Child* 2020;**105**:996-998.
7. Hamer HM, Mulder AHL, de Boer NK, et al. Impact of preanalytical factors on calprotectin concentration in stool: a multiassay comparison. *J Appl Lab Med* 2022;**7**:1401-1411.
8. Oyaert M, Van den Bremt S, Boel A, Bossuyt X, Van Hoovels L. Do not forget about pre-analytics in faecal calprotectin measurement! *Clin Chim Acta* 2017;**473**:124-126.
9. Padoan A, D'Incà R, Scapellato ML, et al. Improving IBD diagnosis and monitoring by understanding preanalytical, analytical and biological fecal calprotectin variability. *Clin Chem Lab Med* 2018;**56**:1926-1935.
10. Lasson A, Stotzer PO, Öhman L, Isaksson S, Sapnara M, Strid H. The intra-individual variability of faecal calprotectin: a prospective study in patients with active ulcerative colitis. *J Crohns Colitis* 2015;**9**:26-32.
11. Dhaliwal A, Zeino Z, Tomkins C, et al. Utility of faecal calprotectin in inflammatory bowel disease (IBD): what cut-offs should we apply? *Frontline Gastroenterol* 2014;**6**:14-19.
12. Du L, Foshaug R, Huang VW, et al. Within-stool and within-day sample variability of fecal calprotectin in patients with inflammatory bowel disease: a prospective observational study. *J Clin Gastroenterol* 2018;**52**:235-240.
13. Calafat M, Cabré E, Mañosa M, Lobatón T, Marín L, Domènech E. High within-day variability of fecal calprotectin levels in patients with active ulcerative colitis: what is the best timing for stool sampling? *Inflamm Bowel Dis* 2015;**21**:1072-1076.
14. Kristensen V, Malmstrøm GH, Skar V, Røseth A, Moum B. Clinical importance of faecal calprotectin variability in inflammatory bowel disease: intra-individual variability and standardisation of sampling procedure. *Scand J Gastroenterol* 2016;**51**:548-555.
15. Cremer A, Ku J, Amininejad L, et al. Variability of faecal calprotectin in inflammatory bowel disease patients: an observational case-control study. *J Crohns Colitis* 2019;**13**:1372-1379.
16. Moum B, Jahnsen J, Bernklev T. Fecal calprotectin variability in Crohn's disease. *Inflamm Bowel Dis* 2010;**16**:1091-1092.
17. Naismith GD, Smith LA, Barry SJ, et al. A prospective single-centre evaluation of the intra-individual variability of faecal calprotectin in quiescent Crohn's disease. *Aliment Pharmacol Ther* 2013;**37**:613-621.
18. Walsham NE, Sherwood RA. Fecal calprotectin in inflammatory bowel disease. *Clin Exp Gastroenterol* 2016;**9**:21-29.
19. D'Amico F, Nancey S, Danese S, Peyrin-Biroulet L. A practical guide for faecal calprotectin measurement: myths and realities. *J Crohns Colitis* 2021;**15**:152-161.
20. Juricic G, Brencic T, Tesija-Kuna A, Njegovan M, Honovic L. Faecal calprotectin determination: impact of preanalytical sample treatment and stool consistency on within- and between-method variability. *Biochem Med (Zagreb)* 2019;**29**:010707.
21. Murray J, Kok KB, Ayling RM. Fecal calprotectin in gastrointestinal disease. *Clin Chem* 2023;**69**:699-710.
22. Kapel N, Ouni H, Benahmed NA, Barbot-Trystram L. Fecal calprotectin for the diagnosis and management of inflammatory bowel diseases. *Clin Transl Gastroenterol* 2023;**14**:e00617.
23. Lee JM, Jang JH, Ryu JH, et al. A comparison of diagnostic performance between two quantitative rapid fecal calprotectin assays in detecting active inflammatory bowel disease. *PLoS One* 2021;**16**:e0255974.
24. Acevedo D, Salvador MP, Girbes J, Estan N. Fecal calprotectin: a comparison of two commercial enzymoimmunoassays and study of fecal extract stability at room temperature. *J Clin Med*

Res 2018;**10**:396-404.
25. Whitehead SJ, French J, Brookes MJ, Ford C, Gama R. Between-assay variability of faecal calprotectin enzyme-linked immunosorbent assay kits. *Ann Clin Biochem* 2013;**50**:53-61.
26. Pelkmans LPJ, de Groot MJM, Curvers J. Analytical performance and clinicopathologic correlation of four fecal calprotectin methods. *Am J Clin Pathol* 2019;**152**:392-398.
27. Mandic-Havelka A, Nilsen T, Sunde K, Norell M, Hansson LO, Larsson A. Turbidimetric determination of fecal calprotectin using two table top chemistry analyzers: Mindray BS-200E and Cobas® c111. *Clin Lab* 2017;**63**:907-913.
28. De Sloovere MMW, De Smet D, Baert FJ, Debrabandere J, Vanpoucke HJM. Analytical and diagnostic performance of two automated fecal calprotectin immunoassays for detection of inflammatory bowel disease. *Clin Chem Lab Med* 2017;**55**:1435-1446.
29. Hiraoka S, Takashima S, Inokuchi T, et al. The novel latex agglutination turbidimetric immunoassay system for simultaneous measurements of calprotectin and hemoglobin in feces. *Intest Res* 2019;**17**:202-209.
30. Goll R, Heitmann R, Moe ØK, Carlsen K, Florholmen J. Head to head comparison of two commercial fecal calprotectin kits as predictor of Mayo endoscopic sub-score and mucosal TNF expression in ulcerative colitis. *PLoS One* 2019;**14**:e0224895.
31. Radillo O, Pascolo L, Martelossi S, Dal Bo S, Ventura A. Fecal calprotectin: diagnostic accuracy of the immunochromatographic CalFast assay in a pediatric population. *J Clin Lab Anal* 2016;**30**:500-505.
32. Mirsepasi-Lauridsen HC, Bachmann Holmetoft U, Ingdam Halkjær S, Angeliki Krogfelt K, Munk Petersen A. Comparison of three commercial fecal calprotectin ELISA test kits used in patients with inflammatory bowel disease. *Scand J Gastroenterol* 2016;**51**:211-217.
33. Labaere D, Smismans A, Olmen AV, et al. Comparison of six different calprotectin assays for the assessment of inflammatory bowel disease. *United European Gastroenterol J* 2014;**2**:30-37.
34. Lin L, Wyness SP, Jensen R, et al. Comparison of next-generation assays for fecal calprotectin vs the PhiCal assay. *Am J Clin Pathol* 2022;**157**:252-256.
35. Volkers A, Löwenberg M, Braad M, et al. Validation study of novel point-of-care tests for infliximab, adalimumab and C-reactive protein in capillary blood and calprotectin in faeces in an ambulatory inflammatory bowel disease care setting. *Diagnostics (Basel)* 2023;**13**:1712.
36. Schulz C, Wex T, von Arnim U, Malfertheiner P. Validation of two calprotectin rapid tests in daily routine. *Clin Lab* 2016;**62**: 1249-1254.
37. 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**:207-214.
38. Damms A, Bischoff SC. Validation and clinical significance of a new calprotectin rapid test for the diagnosis of gastrointestinal diseases. *Int J Colorectal Dis* 2008;**23**:985-992.
39. Inoue K, Aomatsu T, Yoden A, Okuhira T, Kaji E, Tamai H. Usefulness of a novel and rapid assay system for fecal calprotectin in pediatric patients with inflammatory bowel diseases. *J Gastroenterol Hepatol* 2014;**29**:1406-1412.
40. Osredkar J, Kurent T, Fabjan T, Kumer K, Božnar Alič E, Drobne D. The comparison of the three assays for determination of fecal calprotectin in inflammatory bowel disease. *Biochem Med (Zagreb)* 2021;**31**:020707.
41. Kolho KL, Turner D, Veereman-Wauters G, et al. Rapid test for fecal calprotectin levels in children with Crohn disease. *J Pediatr Gastroenterol Nutr* 2012;**55**:436-439.
42. Walker GJ, Chanchlani N, Thomas A, et al. Primary care faecal calprotectin testing in children with suspected inflammatory bowel disease: a diagnostic accuracy study. *Arch Dis Child* 2020;**105**:957-963.
43. 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**:825-831.
44. Vinding KK, Elsberg H, Thorkilgaard T, et al. Fecal calprotectin measured by patients at home using smartphones—a new clinical tool in monitoring patients with inflammatory bowel disease. *Inflamm Bowel Dis* 2016;**22**:336-344.
45. Haisma SM, Galaurchi A, Almahwzi S, Adekanmi Balogun JA, Muller Kobold AC, van Rheenen PF. Head-to-head comparison of three stool calprotectin tests for home use. *PLoS One* 2019;**14**:e0214751.
46. Hejl J, Theede K, Møllgren B, et al. Point of care testing of fecal calprotectin as a substitute for routine laboratory analysis. *Pract Lab Med* 2018;**10**:10-14.
47. Solitano V, Alfarone L, D'Amico F, Peyrin-Biroulet L, Danese S. IBD goes home: from telemedicine to self-administered advanced therapies. *Expert Opin Biol Ther* 2022;**22**:17-29.
48. Lerchova T, Hradsky O, Copova I, Potuznikova K, Gonsorcikova L, Bronsky J. The accuracy of a home-performed faecal calprotectin test in paediatric patients with inflammatory bowel disease. *J Pediatr Gastroenterol Nutr* 2019;**69**:75-81.
49. Heida A, Knol M, Kobold AM, Bootsman J, Dijkstra G, van Rheenen PF. Agreement between home-based measurement of stool calprotectin and ELISA results for monitoring inflammatory bowel disease activity. *Clin Gastroenterol Hepatol* 2017;**15**:1742-1749.
50. Bello C, Roseth A, Guardiola J, et al. Usability of a home-based test for the measurement of fecal calprotectin in asymptomatic IBD patients. *Dig Liver Dis* 2017;**49**:991-996.
51. Elkjaer M, Burisch J, Voxen Hansen V, Deibjerg Kristensen B, Slott Jensen JK, Munkholm P. A new rapid home test for faecal calprotectin in ulcerative colitis. *Aliment Pharmacol Ther* 2010;**31**:323-330.
52. Pathirana WGW, Chubb SP, Gillett MJ, Vasikaran SD. Faecal calprotectin. *Clin Biochem Rev* 2018;**39**:77-90.
53. Tibble JA, Sigthorsson G, Foster R, Forgacs I, Bjarnason I. Use of surrogate markers of inflammation and Rome criteria to distinguish organic from nonorganic intestinal disease. *Gastroenterology* 2002;**123**:450-460.
54. Shane AL, Mody RK, Crump JA, et al. 2017 Infectious Diseases Society of America clinical practice guidelines for the diagnosis and management of infectious diarrhea. *Clin Infect Dis* 2017;**65**:e45-e80.
55. Dajti E, Frazzoni L, Iascone V, et al. Systematic review with meta-analysis: diagnostic performance of faecal calprotectin in distinguishing inflammatory bowel disease from irritable bowel syndrome in adults. *Aliment Pharmacol Ther* 2023;**58**:1120-1131.
56. von Roon AC, Karamountzos L, Purkayastha S, et al. Diagnostic precision of fecal calprotectin for inflammatory bowel disease and colorectal malignancy. *Am J Gastroenterol* 2007;**102**:803-813.
57. Laserna-Mendieta EJ, Lucendo AJ. Faecal calprotectin in inflammatory bowel diseases: a review focused on meta-analyses and routine usage limitations. *Clin Chem Lab Med* 2019;**57**:1295-1307.
58. 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.
59. Manz M, Burri E, Rothen C, et al. Value of fecal calprotectin in the evaluation of patients with abdominal discomfort: an observational study. *BMC Gastroenterol* 2012;**12**:5.
60. Kopylov U, Yung DE, Engel T, et al. Fecal calprotectin for the prediction of small-bowel Crohn's disease by capsule endoscopy: a systematic review and meta-analysis. *Eur J Gastroenterol Hepatol* 2016;**28**:1137-1144.

61. Arasaradnam RP, Brown S, Forbes A, et al. Guidelines for the investigation of chronic diarrhoea in adults: British Society of Gastroenterology, 3rd edition. *Gut* 2018;**67**:1380-1399.
62. Asiri AS, Algarni SS, Althubaiti AQ, Alzubaidi MA, Alghamdi JA, Almalki GA. Fecal calprotectin and organic gastrointestinal disease: a systematic review. *Cureus* 2023;**15**:e45019.
63. Lozoya Angulo ME, de Las Heras Gómez I, Martinez Villanueva M, Noguera Velasco JA, Avilés Plaza F. Faecal calprotectin, an useful marker in discriminating between inflammatory bowel disease and functional gastrointestinal disorders. *Gastroenterol Hepatol* 2017;**40**:125-131.
64. Jung ES, Lee SP, Kae SH, Kim JH, Kim HS, Jang HJ. Diagnostic accuracy of fecal calprotectin for the detection of small bowel Crohn's disease through capsule endoscopy: an updated meta-analysis and systematic review. *Gut Liver* 2021;**15**:732-741.
65. Menees SB, Powell C, Kurlander J, Goel A, Chey WD. A meta-analysis of the utility of C-reactive protein, erythrocyte sedimentation rate, fecal calprotectin, and fecal lactoferrin to exclude inflammatory bowel disease in adults with IBS. *Am J Gastroenterol* 2015;**110**:444-454.
66. An YK, Prince D, Gardiner F, et al. Faecal calprotectin testing for identifying patients with organic gastrointestinal disease: systematic review and meta-analysis. *Med J Aust* 2019;**211**:461-467.
67. Shi JT, Chen N, Xu J, et al. Diagnostic accuracy of fecal calprotectin for predicting relapse in inflammatory bowel disease: a meta-analysis. *J Clin Med* 2023;**12**:1206.
68. Colombel JF, Panaccione R, Bossuyt P, et al. Effect of tight control management on Crohn' disease (CALM): a multicentre, randomised, controlled phase 3 trial. *Lancet* 2017;**390**:2779-2789.
69. Danese S, Vermeire S, D'Haens G, et al; STARDUST study group. Treat to target versus standard of care for patients with Crohn's disease treated with ustekinumab (STARDUST): an open-label, multicentre, randomised phase 3b trial. *Lancet Gastroenterol Hepatol* 2022;**7**:294-306.
70. Lee SH, Kim MJ, Chang K, et al. Fecal calprotectin predicts complete mucosal healing and better correlates with the ulcerative colitis endoscopic index of severity than with the Mayo endoscopic subscore in patients with ulcerative colitis. *BMC Gastroenterol* 2017;**17**:110.
71. Theede K, Holck S, Ibsen P, Ladelund S, Nordgaard-Lassen I, Nielsen AM. Level of fecal calprotectin correlates with endoscopic and histologic inflammation and identifies patients with mucosal healing in ulcerative colitis. *Clin Gastroenterol Hepatol* 2015;**13**:1929-1936.e1.
72. Cannatelli R, Bazarova A, Zardo D, et al. Fecal calprotectin thresholds to predict endoscopic remission using advanced optical enhancement techniques and histological remission in IBD patients. *Inflamm Bowel Dis* 2021;**27**:647-654.
73. Bohra A, Mohamed G, Vasudevan A, Lewis D, Van Langenberg DR, Segal JP. The utility of faecal calprotectin, lactoferrin and other faecal biomarkers in discriminating endoscopic activity in Crohn's disease: a systematic review and meta-analysis. *Biomedicines* 2023;**11**:1408.
74. Yzet C, Brazier F, Hautefeuille V, et al. Non-invasive evaluation of mucosal healing by intestinal ultrasound or fecal calprotectin is efficient in Crohn's disease: a cross-sectional study. *Clin Res Hepatol Gastroenterol* 2024;**48**:102387.
75. Turvill JL, Turnock D, Cottingham D, et al. The Fast Track FIT study: diagnostic accuracy of faecal immunochemical test for haemoglobin in patients with suspected colorectal cancer. *Br J Gen Pract* 2021;**71**:e643-e651.
76. Dulai PS, Singh S, Jairath V, et al. Prevalence of endoscopic improvement and remission according to patient-reported outcomes in ulcerative colitis. *Aliment Pharmacol Ther* 2020;**51**:435-445.
77. Dulai PS, Battat R, Barsky M, et al. Incorporating fecal calprotectin into clinical practice for patients with moderate-to-severely active ulcerative colitis treated with biologics or small-molecule inhibitors. *Am J Gastroenterol* 2020;**115**:885-894.
78. Adamina M, Minozzi S, Warusavitarne J, et al. ECCO guidelines on therapeutics in Crohn's disease: surgical treatment. *J Crohns Colitis* 2024;**18**:1556-1582.
79. Wright EK, Kamm MA, De Cruz P, et al. Comparison of fecal inflammatory markers in Crohn's disease. *Inflamm Bowel Dis* 2016;**22**:1086-1094.
80. Wright EK, Kamm MA, De Cruz P, et al. Measurement of fecal calprotectin improves monitoring and detection of recurrence of Crohn's disease after surgery. *Gastroenterology* 2015;**148**:938-947.e1.
81. Boschetti G, Laidet M, Moussata D, et al. Levels of fecal calprotectin are associated with the severity of postoperative endoscopic recurrence in asymptomatic patients with Crohn's disease. *Am J Gastroenterol* 2015;**110**:865-872.
82. Boube M, Laharie D, Nancey S, et al; POPCUR study group. Variation of faecal calprotectin level within the first three months after bowel resection is predictive of endoscopic postoperative recurrence in Crohn's disease. *Dig Liver Dis* 2020;**52**:740-744.
83. Qiu Y, Mao R, Chen BL, et al. Fecal calprotectin for evaluating postoperative recurrence of Crohn's disease: a meta-analysis of prospective studies. *Inflamm Bowel Dis* 2015;**21**:315-322.
84. Tham YS, Yung DE, Fay S, et al. Fecal calprotectin for detection of postoperative endoscopic recurrence in Crohn's disease: systematic review and meta-analysis. *Therap Adv Gastroenterol* 2018;**11**:1756284818785571.
85. Ardalan ZS, Friedman AB, Con D, et al. Accuracy of gastrointestinal ultrasound and calprotectin in the assessment of inflammation and its location in patients with an ileoanal pouch. *J Crohns Colitis* 2022;**16**:79-90.
86. Farkas K, Bálint A, Bor R, et al. Faecal matrix metalloprotease-9 is a more sensitive marker for diagnosing pouchitis than faecal calprotectin: results from a pilot study. *Expert Rev Gastroenterol Hepatol* 2015;**9**:387-392.
87. Uchino M, Horio Y, Kuwahara R, Kusunoki K, Nagano K, Ikeuchi H. Associations between pouchitis and fecal calprotectin after restorative proctocolectomy in patients with ulcerative colitis. *Inflamm Intest Dis* 2023;**8**:77-83.
88. McKechnie T, Lee Y, Kruse C, et al. The role of fecal calprotectin in the diagnosis of acute pouchitis following IPAA for ulcerative colitis: a systematic clinical review. *Int J Colorectal Dis* 2020;**35**:1619-1628.
89. Falloon K, Cohen BL, Ottichilo R, Grove D, Rieder F, Qazi T. Biomarkers for the evaluation of pouch inflammation: a systematic review. *Crohns Colitis 360* 2022;**4**:otac043.
90. Yamamoto T, Shimoyama T, Bamba T, Matsumoto K. Consecutive monitoring of fecal calprotectin and lactoferrin for the early diagnosis and prediction of pouchitis after restorative proctocolectomy for ulcerative colitis. *Am J Gastroenterol* 2015;**110**:881-887.
91. Jacques P, Elewaut D. Spondyloarthritis: Gut inflammation in SpA: flagellin joins the dance? *Nat Rev Rheumatol* 2014;**10**:5-6.
92. Fauny M, D'Amico F, Bonovas S, et al. Faecal calprotectin for the diagnosis of bowel inflammation in patients with rheumatological diseases: a systematic review. *J Crohns Colitis* 2020;**14**:688-693.
93. Badran S, Saweres A, GamalEldeen H, et al. Screening of silent inflammatory bowel disease in seronegative spondyloarthritis patients: fecal calprotectin, ileo-colonoscopy and histopathology assessment. *Egypt Rheumatol* 2024;**46**:6-11.
94. Di Brizzi EV, Rocco A, Babino G, Buononato D, Argenziano G, Balato A. Evaluation of the role of faecal calprotectin in the management of psoriatic patients under treatment with biologic drugs. *Biomedicines* 2022;**10**:2968.

95. Onac IA, Clarke BD, Tacu C, et al. Secukinumab as a potential trigger of inflammatory bowel disease in ankylosing spondylitis or psoriatic arthritis patients. *Rheumatology (Oxford)* 2021;**60**: 5233-5238.
96. Ma Y, Fan D, Xu S, et al. Calprotectin in spondyloarthritis: a systematic review and meta-analysis. *Int Immunopharmacol* 2020;**88**:106948.
97. Klingberg E, Strid H, Ståhl A, et al. A longitudinal study of fecal calprotectin and the development of inflammatory bowel disease in ankylosing spondylitis. *Arthritis Res Ther* 2017;**19**:21.
98. Truong SL, Chin J, Liew DFL, et al. Systematic review and meta-analysis of inflammatory bowel disease adverse events with anti-interleukin 17A agents and tumor necrosis factor inhibitors in rheumatic disease and skin psoriasis. *Rheumatol Ther* 2021;**8**:1603-1616.
99. Zhao SS, Harrison SR, Thompson B, et al. The 2025 British Society for Rheumatology guideline for the treatment of axial spondyloarthritis with biologic and targeted synthetic DMARDs. *Rheumatology (Oxford)* 2025;**64**:3242-3254.
100. Julsgaard M, Hvas CL, Gearry RB, et al. Fecal calprotectin is not affected by pregnancy: clinical implications for the management of pregnant patients with inflammatory bowel disease. Inflamm Bowel Dis 2017;23:1240-1246.
101. Doherty J, Moore RL, Kivlehan C, et al. Trends in faecal calprotectin levels during pregnancy in non-IBD patients. *Ir Med J* 2022;**115**:692.
102. Kim ES, Tarassishin L, Eisele C, et al; Mount Sinai Road to Prevention Study Group. Longitudinal changes in fecal calprotectin levels among pregnant women with and without inflammatory bowel disease and their babies. *Gastroenterology* 2021;**160**:1118-1130.
103. Tandon P, Leung K, Yusuf A, Huang VW. Noninvasive methods for assessing inflammatory bowel disease activity in pregnancy: a systematic review. *J Clin Gastroenterol* 2019;**53**:574-581.
104. Rottenstreich A, Mishael T, Granovsky SG, et al. Clinical utility of fecal calprotectin in monitoring disease activity and predicting relapse in pregnant patients with inflammatory bowel diseases. *Eur J Intern Med* 2020;**77**:105-110.
105. Al-Jabri R, Wetwittayakhlang P, Lakatos PL. Monitoring of inflammatory bowel disease in pregnancy: a review of the different modalities. *J Clin Med* 2023;**12**:7343.
106. Davidson F, Lock RJ. Paediatric reference ranges for faecal calprotectin: a UK study. *Ann Clin Biochem* 2017;**54**:214-218.
107. Song JY, Lee YM, Choi YJ, Jeong SJ. Fecal calprotectin level in healthy children aged less than 4 years in South Korea. *J Clin Lab Anal* 2017;**31**:e22113.
108. Li F, Ma J, Geng S, et al. Fecal calprotectin concentrations in healthy children aged 1-18 months. *PLoS One* 2015;**10**:e0119574.
109. Roca M, Rodriguez Varela A, Donat E, et al. Fecal calprotectin and eosinophil-derived neurotoxin in healthy children between 0 and 12 years. *J Pediatr Gastroenterol Nutr* 2017;**65**:394-398.
110. Roca M, Rodriguez Varela A, Carvajal E, et al. Fecal calprotectin in healthy children aged 4-16 years. *Sci Rep* 2020;**10**:20565.
111. Kolho KL, Alfthan H. Concentration of fecal calprotectin in 11,255 children aged 0-18 years. *Scand J Gastroenterol* 2020;**55**:1024-1027.
112. Holtman GA, Lisman-van Leeuwen Y, Day AS, et al. Use of laboratory markers in addition to symptoms for diagnosis of inflammatory bowel disease in children: a meta-analysis of individual patient data. *JAMA Pediatr* 2017;**171**:984-991.
113. Degraeuwe PL, Beld MP, Ashorn M, et al. Faecal calprotectin in suspected paediatric inflammatory bowel disease. *J Pediatr Gastroenterol Nutr* 2015;**60**:339-346.
114. Sipponen T, Kolho KL. Faecal calprotectin in children with clinically quiescent inflammatory bowel disease. *Scand J Gastroenterol* 2010;**45**:872-877.
115. Molander P, Färkkilä M, Ristimäki A, et al. Does fecal calprotectin predict short-term relapse after stopping TNFα-blocking agents in inflammatory bowel disease patients in deep remission? *J Crohns Colitis* 2015;**9**:33-40.
116. Walkiewicz D, Werlin SL, Fish D, Scanlon M, Hanaway P, Kugathasan S. Fecal calprotectin is useful in predicting disease relapse in pediatric inflammatory bowel disease. *Inflamm Bowel Dis* 2008;**14**:669-673.
117. Sýkora J, Siala K, Huml M, Varvařovská J, Schwarz J, Pomahačová R. Evaluation of faecal calprotectin as a valuable non-invasive marker in distinguishing gut pathogens in young children with acute gastroenteritis. *Acta Paediatr* 2010;**99**: 1389-1395.
118. Duman M, Gencpinar P, Biçmen M, et al. Fecal calprotectin: can be used to distinguish between bacterial and viral gastroenteritis in children? *Am J Emerg Med* 2015;**33**:1436-1439.
119. Mitrogiorgou M, Karalexi MA, Papaevangelou V, et al. Faecal calprotectin: a useful biomarker in paediatric juvenile polyps. *J Paediatr Child Health* 2020;**56**:1824-1826.
120. Olafsdottir I, Nemeth A, Lörinc E, Toth E, Agardh D. Value of fecal calprotectin as a biomarker for juvenile polyps in children investigated with colonoscopy. *J Pediatr Gastroenterol Nutr* 2016;**62**:43-46.
121. Biskou O, Gardner-Medwin J, Mackinder M, et al. Faecal calprotectin in treated and untreated children with coeliac disease and juvenile idiopathic arthritis. *J Pediatr Gastroenterol Nutr* 2016;**63**:e112-e115.
122. Rajani S, Huynh HQ, Shirton L, et al. A Canadian study toward changing local practice in the diagnosis of pediatric celiac disease. *Can J Gastroenterol Hepatol* 2016;**2016**:6234160.