RESEARCH

Validation of a care pathway for the use of faecal calprotectin in monitoring patients with Crohn's disease

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ABSTRACT

Introduction We have previously published an evidence-based care pathway for the use of faecal calprotectin (FC) to monitor patients with Crohn's disease established on therapy. Patients are treated as low, intermediate or high risk of continuing Crohn's disease activity based on their FC, whatever their phenotype and surgical status are. Low-risk patients (FC <100 µg/g) are offered 12 monthly follow-ups or step down of therapy if asymptomatic or initial expectant symptomatic treatment. Intermediate-risk patients (FC 100–250 µg/g) are reviewed at 6 months with a repeat FC. High-risk patients (two consecutive FCs >250 µg/g) are flagged up to the responsible clinician as likely having an active Crohn's disease.

Methods To validate this care pathway over a 2-year period, by determining its negative predictive value (NPV) and positive predictive value (PPV).

Results 123 patients were managed by means of the care pathway for a mean of 24.4 months. The NPV and PPV were 0.97 (CI 0.93 to 0.98) and 0.85 (CI 0.80 to 0.94), respectively (sensitivity: 0.92 (0.83 to 0.96) and specificity: 0.95 (0.92 to 0.98)). Importantly 69% of patients with FC >250 µg/g were in clinical remission, the care pathway identifying patients who would benefit from presymptomatic disease modification.

Conclusions This validation of a pragmatic clinical care pathway demonstrates a safe and effective mechanism by which to use FC to monitor risk of disease activity in patients with Crohn's disease established on therapy. It provides a framework for prioritising follow-up and for identifying patients at risk of continuing disease activity or those in whom therapy could be stepped down.

INTRODUCTION

Faecal calprotectin (FC) is now widely used to inform clinical decision making in patients with Crohn's disease.^{1–7} Its precise role however remains uncertain in terms of the FC value cut-offs that should be set and the timelines that should support Crohn's disease activity monitoring.⁸ We have previously published an evidence-based care pathway for the use of FC to monitor patients with Crohn's disease established on therapy (figure 1).⁹ It is designed to assist the clinician in four key scenarios:^{10–11}

1. in assessing those patients who are asymptomatic but in whom it is not known whether mucosal healing has been achieved. Identification of this cohort of patients may allow time for the optimisation of therapy and so prevent future symptomatic flare of disease;^{1–2 12–13}
2. in distinguishing symptoms caused by an active Crohn's disease from those of a superimposed irritable bowel syndrome (IBS);^{14–15}
3. in identifying those patients with complete mucosal healing in whom the step down of medical therapy could be considered;^{12–16–21}
4. in optimising clinic management and in tailoring outpatient follow-up to the needs of each patient.^{22–23} Here, FC permits a risk assessment of likelihood of future Crohn's disease activity. Outpatient scheduling could be optimised by being based on Crohn's disease activity risk.

The care pathway is applied to all Crohn's disease phenotypes and to post-surgical patients but only once patients are established on maintenance therapy. This includes steroids only if used in the

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Figure 1 Care pathway for the use of faecal calprotectin in the management of patients with Crohn's disease established on therapy.

context of a long-term maintenance therapy. Patients taking infliximab, adalimumab or methotrexate are judged to be established on therapy after 2 months, while for azathioprine and 6-mercaptopurine it is 4 months. Monitoring of postsurgical patients begins at 3 months.

The care pathway uses a 'traffic light' system of assessment based on the following cut-offs: FC <100 µg/g, FC 100–250 µg/g and FC >250 µg/g.

Faecal calprotectin <100 µg/g

Here, quiescent Crohn's disease is likely and the likelihood of a relapse or recurrence in the next 12 months is low.⁹ If continued therapy is indicated and the patient is in clinical remission, outpatient follow-up is scheduled for 12 months. Onset of lower gastrointestinal symptoms during this time is treated in the first instance as likely IBS in nature. Persisting symptoms thereafter prompt a repeat FC and a conventional clinical assessment. Only patients with FC <100 µg/g are considered for the step down of therapy, this being informed by thiopurine metabolites and tumour necrosis factor (TNF)α antibodies and anti-antibodies as appropriate. The step down of therapy or

postoperative patients is then monitored with three monthly FCs in the first 12 months.

Faecal calprotectin 100–250 µg/g

Patients with FC between 100 and 250 µg/g are likely to have either quiescent Crohn's disease or low-grade activity.⁹ Patients are unlikely to suffer a symptomatic flare or relapse in the next 6 months and so follow-up is scheduled for this time where a further FC is requested. As with those patients with FC <100, the new onset of symptoms during this period is likely IBS in nature and initial expectant treatment is offered before further review.

Faecal calprotectin >250 µg/g

In those with FC >250 µg/g, the test is repeated.²⁴ A second result >250 µg/g suggests that an active Crohn's disease is likely and here the responsible clinician is prompted to reassess the patient.⁹ In practice, this takes the form of investigation if the patient is asymptomatic, the mode of investigation being determined on clinical grounds. If the patient has remained symptomatic since being established on maintenance therapy, the clinician may decide to reinvestigate or simply to escalate therapy (informed by thiopurine metabolites and TNFα

antibodies and anti-antibodies as appropriate). Once a new management plan has been established, the patient re-enters the FC care pathway.

AIMS

We undertook to complete the audit cycle by validating the care pathway prospectively in the setting of a single secondary care gastroenterology service over a 2-year period.

METHODS

The evaluation was registered with the Clinical Effectiveness Unit at the York Teaching Hospital NHS Foundation Trust as an audit of the care pathway (number 2653).

From January 2014, patients with Crohn's disease once established on therapy were entered into the care pathway (figure 1). A secure spreadsheet was developed for data input. Crohn's disease phenotype, previous surgery, patient symptomatology and maintenance therapy were documented.²⁵ As the care pathway was implemented for each patient, FC results, interventions and clinical outcomes were recorded until April 2016.

Because this evaluation occurred in the context of a conventional clinical practice and any symptomatology was to be recorded, no formal symptom-based Crohn's disease activity index was indicated. Patient symptoms and well-being were recorded as reported in clinic correspondence. Endoscopic and histological and radiological findings were recorded as reported by the responsible clinician. Similarly, while these were descriptive rather than being formally defined, they were judged sufficient for the evaluation since mucosal healing per se rather than degrees of disease activity was the measured standard.

Assay

Stool samples were processed by the Department of Clinical Biochemistry at the York Hospital. The samples were stored at $<4^{\circ}\text{C}$ prior to extraction (weighing method). The extracts were stored at -20°C and then analysed in batches. The stool extract was analysed using a monoclonal ELISA (EK-CAL calprotectin ELISA, Buhlmann, supplied by Alpha Laboratories) to determine the FC level. The normal cut-off was taken to be $<50\text{ }\mu\text{g/g}$ in line with the manufacturer's guidance. A quality control sample set at a level of 150 was present in every test batch, the coefficient of variation being 5%. The upper limit of linearity for the assay was $600\text{ }\mu\text{g/g}$.

Analysis

The primary analysis of the care pathway evaluation was *any* evidence of Crohn's disease activity relative to the FC $250\text{ }\mu\text{g/g}$ cut-off. However, since the care pathway acts as a risk assessment tool, it is *predictive* of future quiescent disease when FC is $\leq 250\text{ }\mu\text{g/g}$

while it is *indicative* of an active Crohn's disease if FC is $>250\text{ }\mu\text{g/g}$ when repeated. Therefore, the measure for the former was the proportion of patients remaining free of an active Crohn's disease until the next point of review and, for the latter, it was the proportion of patients with an active Crohn's disease at that time. Since the care pathway is intended for a heterogeneous, that is, clinically relevant group of patients, the outcome measure of *any* evidence of Crohn's disease activity was derived from a composite of endoscopic, radiological findings and patient symptomatology in the subsequent 3-month, 6-month or 12-month period of follow-up as the care pathway directed. It was recognised in the context of this non-trial evaluation that to investigate all clinically asymptomatic patients with FC $<100\text{ }\mu\text{g/g}$ would be inappropriate. To offset this, it was judged that the 2-year evaluation cycle would be sufficient to allow any occult active disease to become manifest.²⁴

The negative predictive value (NPV) and positive predictive value (PPV), sensitivity and specificity for the FC cut-off $>250\text{ }\mu\text{g/g}$ were determined. A receiver-operator characteristic (ROC) curve was generated. No comparator data are presented, the care pathway itself being validated.

RESULTS

Three hundred and thirty care pathway interventions, involving 123 patients, took place over a mean of 24.4 months (SD 6.9) for each patient. A further nine patients were not prepared to provide stool samples. The distribution of FC during the evaluation was:

- 38.0% FC was $<100\text{ }\mu\text{g/g}$ (low risk),
- 30.5% FC was $100\text{--}2500\text{ }\mu\text{g/g}$ (intermediate risk),
- 31.5% FC was $>2500\text{ }\mu\text{g/g}$ (high risk).

The Montreal classification of disease, maintenance therapy and previous surgery for Crohn's disease for the patients is presented in table 1.

Negative and positive predictive value

The care pathway had a PPV of 0.85 (CI 0.80 to 0.94) and a NPV of 0.97 (CI 0.93 to 0.98), a sensitivity of 0.92 (CI 0.83 to 0.96) and a specificity of 0.95 (CI 0.92 to 0.98). The data are presented as an ROC curve (figure 2).

On 89 occasions, patients' Crohn's disease activity was formally confirmed, endoscopically or radiologically, rather than using the composite that included clinical symptomatology. Here, the care pathway had a PPV of 0.90 (CI 0.80 to 0.95) and a NPV of 0.85 (0.61 to 0.96), a sensitivity of 0.95 (CI 0.86 to 0.99) and a specificity of 0.71 (0.49 to 0.87).

Patients with FC $\leq 250\text{ }\mu\text{g/g}$ (low or intermediate risk)

Eighty three per cent of patients with FC $\leq 250\text{ }\mu\text{g/g}$ were clinically asymptomatic and so for the most part the care pathway supported the existing management plan. For those with FC $<100\text{ }\mu\text{g/g}$, the NPV for an

Table 1 Crohn's disease phenotype, medical therapy and previous surgery

Crohn's disease	Number	Per cent
Montreal classification		
Age at diagnosis		
A1	12	10
A2	79	64
A3	32	26
Location		
L1	44	36
L2	28	23
L3	47	38
L4	7	6
Behaviour		
B1	45	36
B2	62	51
B3	16	13
Perianal disease		
Present	10	8
Therapy		
Nil	24	20
Mesalazine	27	22
Azathioprine	40	33
6-Mercaptopurine	15	12
Methotrexate	10	8
Infliximab	22	18
Adalimumab	23	19
Dual therapy	22	18
Steroid therapy	1	1
Previous surgery		
Right hemicolectomy	44	36
Colectomy	10	8
Small bowel resection	9	7

active Crohn's disease was 0.98 (CI 0.92 to 0.99) and the PPV was 0.47 (CI 0.39 to 0.54). This therefore provided a significant reassurance for the clinician and patient of likely quiescent disease looking ahead for the next 12 months. Here, FC supported an effective trial of IBS therapy in nine patients who became symptomatic and prompted a step down of therapy in six.

False negative patients included flares of perianal disease in the absence of an active luminal disease, a fibrotic stricture, the development of antibodies to biological therapy and a flare after the step down of therapy.

Faecal calprotectin >250 µg/g (high risk)

Sixty nine per cent of patients with FC remaining >250 µg/g when repeated were clinically asymptomatic at the time. This importantly provided the clinician an opportunity to intervene and re-evaluate, prior to the patient suffering a symptomatic flare. Here, escalation of therapy with a subsequent

normalisation of FC (<250 µg/g) was achieved without the patient becoming symptomatic in 56%.

False positives did occur in 6% of cases where FC was >600 µg/g and in 22% of cases where FC was 250–600 µg/g. They tended to be more common in those with colitis, however no significant differences were identified in surgical or non-surgical patients and across the phenotypes. In patients with an active Crohn's disease, 51% had an FC of >600 µg/g, 22% had an FC of 401–600 µg/g and 27% had an FC of 250–400 µg/g.

DISCUSSION

Having previously demonstrated an association between the FC level and disease activity and between the FC level and the risk of subsequent symptomatic flare, we designed a pragmatic care pathway to facilitate in the management of patients with Crohn's disease established on therapy.⁹ This introduced a traffic light system of risk and review, the aim being neither to miss patients with an active disease nor to overinvestigate those in clinical remission. In this way, the cut-off to support likely quiescent Crohn's disease activity (<100 µg/g) was set differently from that suggesting likely an active disease (>250 µg/g repeated). Those patients in the 'amber' or intermediate range between these two cut-offs were followed up more closely. The advantage of the care pathway is that it is simple and workable in supporting clinical decision making, being applied to all Crohn's disease phenotypes and to postsurgical patients since mucosal disease is common to all.²⁶ It is applied only once patients are established on maintenance therapy, whether the patient is symptomatic or not.

Based therefore on the premise that the clinician is aiming to achieve mucosal healing, the care pathway acts as a risk assessment tool. The purpose of this study has been to validate its safety and efficacy.

The validation has been undertaken as an audit and deliberately does not look to address questions that can only be answered within a clinical trial. It does not have a control and so there is no certainty that the care pathway has actually improved clinical outcomes beyond standard care. Neither is there a cost/benefit analysis. The care pathway does not look to correlate the FC levels with degrees of Crohn's disease activity, so there is no need for formal symptom, endoscopic or radiological activity scores. Instead, the evaluation has recorded *any* evidence of Crohn's disease activity as the outcome measure. Recognising that it would be inappropriate to investigate all patients in clinical remission with FC <100 µg/g, we judged that the duration of the study would allow for a true clinical outcome to emerge in those who did not undergo formal investigation.

This study clearly demonstrates that FC can be used within this care pathway as an effective risk assessment tool for patients. Indeed, the FC cut-off values

Figure 2 Receiver-operator characteristic curve (ROC) for faecal calprotectin levels predicting Crohn's disease activity within the care pathway.

used are similar to those proposed in many, largely retrospective or observational studies of such markers of disease activity as the Crohn's Disease Activity Index, endoscopic and radiological findings.^{10 12 27–33} However, none of those have been validated prospectively when applied within a care pathway.

An FC ≤ 250 $\mu\text{g/g}$ can reassure the patient for a defined period of time, support therapeutic decision making in the multidisciplinary team meetings, identify those in whom to consider the step down of therapy and allow for the improved management of clinics. FC supports but does not replace clinical judgement in the context of symptom interpretation. Not surprisingly, FC could not predict for the development of perianal disease activity and problems such as fibrotic strictures and biological antibody formation reflect its limitations. Importantly, previous surgery did not affect the utility of the care pathway.²⁷

Importantly, an FC repeatedly >250 $\mu\text{g/g}$ had a high PPV for an active Crohn's disease. It is particularly important for identifying asymptomatic patients who have occult active Crohn's disease. Here, the care pathway acts as an effective screening tool. This applied to over two-thirds of patients with FC >250 $\mu\text{g/g}$ and provided an opportunity for escalation of therapy before the need for salvage. For most patients with an active Crohn's disease, the FC was repeatedly at the upper limit of the assay (>600 $\mu\text{g/g}$). But false positives occurred only once in five times when FC was between >250 and <600 $\mu\text{g/g}$ (there was a trend for this in patients with pancolitis). We have concluded that the costs of overinvestigating this small cohort are outweighed by the benefits of early disease detection.

Experientially we have found the care pathway to be a particularly useful framework by which to manage patients in a busy outpatient setting. It rapidly became incorporated into clinical practice and was particularly championed by our inflammatory bowel disease nurse specialists who run telephone/virtual

clinics. Since it acts as a risk assessment tool, it permits clinics slots to be set on a needs basis and stool samples can be sent through at allocated times prior to any outpatient review.

In conclusion, we have validated a workable, pragmatic care pathway to support in the ongoing management of patients with Crohn's disease. It is safe and effective. It gives an informed non-invasive structure for prioritising follow-up and for identifying patients at risk of continuing disease activity or those in whom therapy could be stepped down. Despite its audit design, the PPV of 0.85 and a NPV of 0.97 of this clinical validation are compelling and we would encourage its further evaluation in other units.

Significance of this study

What is already known on this topic?

Faecal calprotectin levels correlate with Crohn's disease activity.

What this study adds?

In patients with Crohn's disease, established on therapy, faecal calprotectin effectively identifies those who are likely to remain in remission and those who are likely to flare.

How might it impact on clinical practice in the foreseeable future?

Faecal calprotectin use provides a framework for the safe, efficient management of patients with Crohn's disease, established on therapy.

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REFERENCES

- 1 Neurath MF, Travis SP. Mucosal healing in inflammatory bowel diseases: a systematic review. *Gut* 2012;61:1619–35.
- 2 Desai D, Faubion WA, Sandborn WJ. Review article: biological activity markers in inflammatory bowel disease. *Aliment Pharmacol Ther* 2007;25:247–55.
- 3 Fagerhol MK, Dale I, Anderson I. Release and quantification of leukocyte derived protein (L1). *Scand J Haematol* 1980;24:393–8.
- 4 Tibble JA, Bjarnason I. Non-invasive investigations of inflammatory bowel disease. *World J Gastroenterol* 2001;7:460–5.
- 5 Tibble JA, Teahon K, Thjodleifsson B, *et al.* A simple method for assessing intestinal inflammation in Crohn's disease. *Gut* 2000;47:506–13.

- 6 Lamb CA, Mohiuddin MK, Gicquel J, *et al.* Faecal calprotectin or lactoferrin can identify postoperative recurrence in Crohn's disease. *Br J Surg* 2009;96:663–74.
- 7 Wagner M, Peterson CG, Ridefelt P, *et al.* Fecal markers of inflammation used as surrogate markers for treatment outcome in relapsing inflammatory bowel disease. *World J Gastroenterol* 2008;14:5584–9; discussion 5588.
- 8 Wright EK. Calprotectin or lactoferrin: do they help? *Dig Dis* 2016;34:98–104.
- 9 Turvill JL. Mapping of Crohn's disease outcomes to faecal calprotectin levels in patients maintained on biologic therapy. *Frontline Gastroenterol* 2014;5:167–75.
- 10 Ferreira-Iglesias R, Barreiro-de Acosta M, Otero Santiago M, *et al.* Fecal calprotectin as predictor of relapse in patients with inflammatory bowel disease under maintenance infliximab therapy. *J Clin Gastroenterol* 2016;50:147–51.
- 11 Peyrin-Biroulet L, Sandborn WJ, Sands BE, *et al.* Selecting Therapeutic Targets in Inflammatory Bowel Disease (STRIDE): determining therapeutic goals for treat-to-target. *Am J Gastroenterol* 2015;110:1324–38.
- 12 Mooiweer E, Severs M, Schipper ME, *et al.* Low fecal calprotectin predicts sustained clinical remission in inflammatory bowel disease patients: a plea for deep remission. *J Crohns Colitis* 2015;9:50–5.
- 13 Molander P, af Björkstén CG, Mustonen H, *et al.* Fecal calprotectin concentration predicts outcome in inflammatory bowel disease after induction therapy with TNF α blocking agents. *Inflamm Bowel Dis* 2012;18:2011–17.
- 14 Diederik K, Hoekman DR, Hummel TZ, *et al.* The prevalence of irritable bowel syndrome-type symptoms in paediatric inflammatory bowel disease, and the relationship with biochemical markers of disease activity. *Aliment Pharmacol Ther* 2016;44:181–8.
- 15 Gracie DJ, Williams CJ, Sood R, *et al.* Poor correlation between clinical disease activity and mucosal inflammation, and the role of psychological comorbidity, in inflammatory bowel disease. *Am J Gastroenterol* 2016;111:541–51.
- 16 Brooks AJ, Sebastian S, Cross SS, *et al.* Outcome of elective withdrawal of anti-tumour necrosis factor- α therapy in patients with Crohn's disease in established remission. *J Crohns Colitis* 2015. doi: 10.1016/j.crohns.2014.09.007. [Epub ahead of print 18 May 2015]
- 17 Kennedy NA, Warner B, Johnston EL. Relapse after withdrawal from anti-TNF therapy for inflammatory bowel disease: an observational study, plus systematic review and meta-analysis. *Aliment Pharmacol Ther* 2016. doi: 10.1111/apt.13547. [Epub ahead of print 19 Feb 2016]
- 18 Pittet V, Froehlich F, Maillard MH, *et al.*, EPACT-II Update Panellists. When do we dare to stop biological or immunomodulatory therapy for Crohn's disease? Results of a multidisciplinary European expert panel. *J Crohns Colitis* 2013;7:820–6.
- 19 Laharie D, Mesli S, El Hajbi F, *et al.* Prediction of Crohn's disease relapse with faecal calprotectin in infliximab responders: a prospective study. *Aliment Pharmacol Ther* 2011;34:462–9.
- 20 Mao R, Xiao YL, Gao X, *et al.* Fecal calprotectin in predicting relapse of inflammatory bowel diseases: a meta-analysis of prospective studies. *Inflamm Bowel Dis* 2012;18:1894–9.
- 21 Louis E, Mary JY, Vernier-Massouille G, *et al.*, Groupe D'études Thérapeutiques Des Affections Inflammatoires Digestives. Maintenance of remission among patients with Crohn's disease on antimetabolite therapy after infliximab therapy is stopped. *Gastroenterology* 2012;142:63–70.
- 22 Mindemark M, Larsson A. Ruling out IBD: estimation of the possible economic effects of pre-endoscopic screening with F-calprotectin. *Clin Biochem* 2012;45:552–5.
- 23 Vinding KK, Elsborg 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;22:336–44.
- 24 Turvill JL. High negative predictive value of a normal faecal calprotectin in patients with symptomatic intestinal disease. *Frontline Gastroenterol* 2012;3:21–8.
- 25 Silverberg MS, Satsangi J, Ahmad T, *et al.* Toward an integrated clinical, molecular and serological classification of inflammatory bowel disease: report of a Working Party of the 2005 Montreal World Congress of Gastroenterology. *Can J Gastroenterol* 2005;19(Suppl A):5–36.
- 26 Herranz Bachiller MT, Barrio Andres J, Fernandez Salazar L, *et al.* The utility of faecal calprotectin to predict post-operative recurrence in Crohn's disease. *Scand J Gastroenterol* 2016;51:720–6.
- 27 Mosli MH, Zou G, Garg SK, *et al.* C-reactive protein, fecal calprotectin, and stool lactoferrin for detection of endoscopic activity in symptomatic inflammatory bowel disease patients: a systematic review and meta-analysis. *Am J Gastroenterol* 2015;110:802–19.
- 28 Cerrillo E, Beltrán B, Pous S, *et al.* Fecal calprotectin in ileal Crohn's disease: relationship with magnetic resonance enterography and a pathology score. *Inflamm Bowel Dis* 2015;21:1572–9.
- 29 Sipponen T, Savilahti E, Kärkkäinen P, *et al.* Fecal calprotectin, lactoferrin, and endoscopic disease activity in monitoring anti-TNF-alpha therapy for Crohn's disease. *Inflamm Bowel Dis* 2008;14:1392–8.
- 30 Scarpa M, D'Incà R, Basso D, *et al.* Fecal lactoferrin and calprotectin after ileocolonic resection for Crohn's disease. *Dis Colon Rectum* 2007;50:861–9.
- 31 Dolwani S, Metzner M, Wassell JJ, *et al.* Diagnostic accuracy of faecal calprotectin estimation in prediction of abnormal small bowel radiology. *Aliment Pharmacol Ther* 2004;20:615–21.
- 32 af Björkstén CG, Nieminen U, Turunen U, *et al.* Surrogate markers and clinical indices, alone or combined, as indicators for endoscopic remission in anti-TNF-treated luminal Crohn's disease. *Scand J Gastroenterol* 2012;47:528–37.
- 33 D'Haens G, Ferrante M, Vermeire S, *et al.* Fecal calprotectin is a surrogate marker for endoscopic lesions in inflammatory bowel disease. *Inflamm Bowel Dis* 2012;18:2218–24.