Faecal calprotectin testing for differentiating amongst inflammatory and non-inflammatory bowel diseases: a systematic review and economic evaluation.

April 26th 2013. (Updated 30th May – highlighting has been removed from some previously academic in confidence sections because the two documents in question have now been published.)

HTA reference number 2012/48

PROSPERO registration CRD 42012003287

Assessment group – Warwick Evidence

Project lead: Norman Waugh

Professor of public health medicine and health technology assessment

Warwick Evidence

Division of Health Sciences

Warwick Medical School

Coventry CV4 7AL

norman.waugh@warwick.ac.uk 02476 151585

Authors;

Prof Norman Waugh

Dr Ewen Cummins, health economist, McMDC Ltd

Dr Pamela Royle, senior research fellow

Dr Ngianga-Bakwin Kandala, principal research fellow

Dr Deepson Shyangdan, research fellow

Dr Ramesh Arasaradnam, consultant gastroenterologist and associate professor

Dr Christine Clar, researcher in systematic reviews

Rhona Johnston, computer analyst, McMDC Ltd

Funding for this study was provided by the Health Technology Assessment programme of the National Institute for Health Research. This project was funded by the HTA programme, on behalf of NICE, as project number 2012/48.

Acknowledgements

We thank the following people for useful advice or information;

Dr Hazel Borthwick

Dr Anjan Dhar*

Dr Paul Henderson

Dr Gurleen Juhti, NICE

Dr Robert Logan*

Barbara Mascialino and Steve Ferguson, Thermofisher Scientific

Dr John O'Malley*

Dr Nick Read*

Dr Simon Whitehead*

Professor David Wilson

The Rotherham group: Dr A Kumar, Dr A Basumani, Prof K Bardhan

*Indicates members of the NICE expert advisory group for this appraisal.

And we thank the following for commenting on sections of a near-final draft of this report;

Dr Robert Logan

Dr Nick Read

Dr Marie Westhouse and Nigel Armstrong, Kleijnen Systematic Reviews, York

Professor David Wilson, University of Edinburgh

Competing interests: none.

Contribution of authors:

Pamela Royle carried out literature searches and led the systematic review of clinical effectiveness

Deepson Shyangdan and Christine Clar assisted with the review of clinical effectiveness

Ngianga-Bakwin Kandala provided statistical support

Ewen Cummins reviewed the economic literature and carried out de novo economic modelling

Rhona Johnston assisted with modelling

Ramesh Arasaradnam provided expert clinical advice and unpublished data

Norman Waugh drafted the report based on contributions from co-authors

Table of Contents

List of Tables and Figures

Tables

Figures

List of abbreviations

ASA	Aminosalicylic acid
ASCA	Anti-Saccharomyces cerevisae antibodies
AUC	Area Under the Curve
BNI	British Nursing Index
BWUS	Bowel wall ultrasonography measurement
BSG	British Society of Gastroenterology
CBT	Cognitive Behavioural Therapy
CD	Crohn's Disease
CDAI	Crohn's disease activity index
CE	Conformité Européenne (European Conformity)
CEAF	Cost-effectiveness acceptability frontier
CEP	Centre for Evidence based Purchasing
CI	Confidence Interval
CRC	Colorectal cancer
CRP	C-reactive protein
CT	Computed tomography
DOR	Diagnostic Odds Ratio
ELISA	Enzyme-linked immunosorbent assay
EQ-5D	EuroQol-5D
ESR	Erythrocyte sedimentation rate
FBC	Full blood count
FC	Faecal calprotectin
FDA	Food and Drug Administration
FN	False Negative
FP	False Positive
GI	Gastrointestinal
GP	General Practitioner
HLA	Human leukocyte antigen
HoDaR	Health Outcomes Data Repository
HRQoL	Health related quality of life
HUI	Health utility index

IBD	Inflammatory bowel disease
IBS	Irritable bowel syndrome
IBS-A	Irritable bowel syndrome – alternating type
IBS-C	Irritable bowel syndrome-constipation
IBS-D	Irritable bowel syndrome-diarrhoea
IBS-M	Irritable bowel syndrome-mixed
ICC	Intraclass correlation coefficient
ICER	Incremental cost-effectiveness ratio
IDDM	Insulin-dependent diabetes mellitus
IP	Intestinal permeability
LR	Likelihood Ratio
MRI	Magnetic resonance imaging
N	Negative
NHS	National Health Service
NICE	National Institute for Health and Clinical Excellence
NLR	Negative Likelihood Ratio
NPV	Negative Predictive Value
NSAIDs	Non-steroidal anti-inflammatory drugs
OBD	Organic bowel disease
pANCA	perinuclear Anti-Neutrophil Cytoplasmic Antibodies
P	Positive
PIBD	Paediatric inflammatory bowel disease
PLR	Positive Likelihood Ratio
POCT	Point of Care Test
PPI	Proton pump inhibitor
PPV	Positive Predictive Value
PSA	Probabilistic sensitivity analysis
QALY	Quality adjusted life year
QoL	Quality of life
QUADAS	Quality Assessment of Diagnostic Accuracy Studies
RCT	Randomised controlled trial
ROC	Receiver operating characteristic
RR	Relative risk

SD	Standard deviation
Se	Sensitivity
SF-36	Short form-36
Sp	Specificity
SROC	Summary receiver operative characteristic
SSRI	Selective serotonin reuptake inhibitor
TA	Technology appraisal
TCA	Tricyclic antidepressant
TN	True negative
TNF	Tumour Necrosis Factor
TP	True Positive
TRFIA	Time-Related Fluorimetric Immunoassay
TSH	Thyroid Stimulating Hormone
TTG	Tissue Transglutaminase (a test for coeliac disease)
TTO	Time trade off
UC	Ulcerative Colitis
UCAI	Ulcerative Colitis Activity Index
YHEC	York Health Economics Consortium

Glossary

Accuracy: Accuracy is the probability that the test yields a correct result (TP+TN)/(P+N)

Bloating: Fullness or swelling in the abdomen that often occurs after meals

Constipation: A condition in which bowel movements are infrequent, hard and dry, and elimination of faeces is difficult and infrequent.

Cost impact: The total cost to the person, the NHS or to society.

Cost-minimisation analysis: A type of economic evaluation used to compare the difference in costs between programs that have the same health outcome.

Cost-utility analysis: A form of cost-effectiveness analysis in which the units of effectiveness are quality-adjusted life-years (QALYs).

Crohn's Disease: A chronic inflammatory disease of the digestive tract that can involve any part of it - from the mouth to the anus. It typically affects the terminal ileum as well as demarcated areas of large bowel, with other areas of the bowel being relatively unaffected. It is often associated with auto-immune disorders outside the bowel, such as rheumatoid arthritis.

Cost-consequences analysis: A type of economic evaluation, whereby both outcomes and costs of alternative interventions are described, without any attempt to combine the results.

Cost effectiveness: The cost per unit of benefit of an intervention. Benefits of different interventions are measured using a single outcome (for example, life-years gained, quality adjusted life-years gained, deaths avoided, heart attacks avoided, cases detected).

Cost-effectiveness analysis: An economic study design in which alternative interventions are compared in terms of cost per unit of effectiveness.

Cost-effectiveness model: An explicit mathematical framework, which is used to represent clinical decision problems and incorporate evidence from a variety of sources in order to estimate the costs and health outcomes.

Cost-of-illness/economic burden studies: An analysis of the total costs incurred by a society due to a specific disease.

Dominance: An intervention is said to be dominated if there is an alternative intervention that is both less costly and more effective

Diagnostic odds ratio: It is defined as the ratio of the odds of the test being positive if the subject has a disease relative to the odds of the test being positive if the subject does not have the disease.

Discounting: Discounting is a method for adjusting the value of costs and outcomes which occur in different time periods into a common time period, usually the present.

Fagan's nomogram: Fagan's nomogram is a graph that uses pre-test probability of IBD and likelihood ratios to estimate the probability of a patient with a positive test having the condition. Examples are shown on pages.

False-negative: Incorrect negative test result – number of diseased persons with a negative test result.

False-positive: Incorrect positive test result – number of non-diseased persons with a positive test result.

Functional Bowel Disorder: In medicine, the term functional bowel disorder refers to a group of disorders which are characterised by chronic abdominal complaints without a structural or biochemical cause that could explain symptoms

Incremental cost-effectiveness ratio: The difference in the mean costs of two interventions in the population of interest divided by the difference in the mean outcomes in the population of interest.

Index test: The test of which performance is being evaluated.

Inflammatory Bowel Disease: General term for any disease characterized by inflammation of the bowel. Two of the most common Inflammatory Bowel Diseases are ulcerative colitis and Crohn's disease

Likelihood ratios: Likelihood ratios (LRs) combine the information from sensitivity and specificity. The LR for a positive test (LR+) is the probability of an individual with IBD having a positive test (sensitivity) divided by the probability of an individual without IBD having a positive test (1 minus specificity).

The LR for a negative test (LR-) is the probability of an individual with IBD having a negative test divided by the probability of an individual without IBD having a negative test.
So LR- = 1 – sensitivity/ specificity.

In those with a positive test, LR+ values of 10 or greater are usually regarded as strong evidence of a disease being present.

Meta-analysis Statistical techniques used to combine the results of two or more studies and obtain a combined estimate of effect.

Negative predictive value: The negative predictive value of a calprotectin test is the probability that a patient with a negative calprotectin test does not have IBD.

Positive predictive value: The positive predictive value is defined as the probability that a patient with a positive calprotectin test has IBD.

Quality adjusted life years (QALYs): An index of survival that is adjusted to account for the patient's quality of life during this time. QALYs have the advantage of incorporating changes in both quantity (longevity/mortality) and quality (morbidity, psychological, functional, social and other factors) of life. Used to measure benefits in cost-utility analysis. The QALYs gained are the mean QALYs associated with one treatment minus the mean QALYs associated with an alternative treatment.

Quality of life: An individual's emotional, social and physical well-being, and his or her ability to perform the ordinary tasks of living.

Receiver operating characteristic curve: A graph that illustrates the trade-offs between sensitivity and specificity, which result from varying the diagnostic threshold.

Reference standard: The best currently available diagnostic test(s), against which the index test is compared.

Sensitivity (of a test): The proportion of individuals classified as positive by the gold (or reference) standard, who are correctly identified by the study test.

Sensitivity analysis: A means of representing uncertainty in the results of economic evaluations. Uncertainty may arise from missing data, imprecise estimates or methodological controversy. Sensitivity analysis also allows for exploring the generalisability of results to other settings. The analysis is repeated using different assumptions to examine the effect on the results.

- One-way simple sensitivity analysis (univariate analysis): each parameter is varied individually inorder to isolate the consequences of each parameter on the results of the study.
- Multi-way simple sensitivity analysis (scenario analysis): two or more parameters are varied at the same time and the overall effect on the results is evaluated.
- Threshold sensitivity analysis: the critical value of parameters above or below which the
- conclusions of the study will change are identified.
- Probabilistic sensitivity analysis: probability distributions are assigned to the uncertain parameters and are incorporated into evaluation models based on decision analytical techniques (For example, Monte Carlo simulation).

Specificity (of a test): The proportion of individuals classified as negative by the gold (or reference) standard, who are correctly identified by the study test.

True-negative: Correct negative test result – number of non-diseases persons with a negative test result.

True-positive: Correct positive test result – number of diseased persons with a positive test result.

Utility: A measure of the strength of an individual's preference for a specific health state in relation to alternative health states. The utility scale assigns numerical values on a scale from 0 (death) to 1 (optimal or 'perfect' health). Health states can be considered worse than death and thus have a negative value.

Visceral hypersensitivity: Enhanced perception or enhanced responsiveness within the gut.

Summary

Lower abdominal symptoms such as pain, diarrhoea and bloating are very common in the population, and are usually due to irritable bowel syndrome (IBS), a troublesome condition that interferes with activities of daily life but one which does not have serious consequences. It is estimated that around 10% of the population will have symptoms suggestive of IBS, though only about half will consult their general practitioners (GPs). IBS is an unexplained bowel disorder, characterised by frequent bouts of bowel disturbance and abdominal discomfort. There is no clear cause, no distinctive pathology and no definitive treatment. Exacerbations may be triggered by diet or stress. Physiological studies often show an increase in bowel sensitivity, and it may be associated with abnormal muscle activity in the wall of the bowel.

The symptoms of IBS may be similar to those of inflammatory bowel disease (IBD), a group of conditions, but comprising mainly Crohn's disease and ulcerative colitis. These are diseases with serious complications, including a high risk of complications requiring surgery, and an increased risk of colorectal cancer.

However the symptoms of IBD can be different in children, many of whom present with non-specific symptoms such as mild abdominal discomfort, lethargy, weight loss or growth impairment. In a large UK and Ireland study, only 25% of children with Crohn's disease presented with the usual triad of diarrhoea, abdominal pain and weight loss. Delays in diagnosis were common, with over a quarter of patients with CD taking over a year to be diagnosed. About 25% of people with IBD, develop it under the age of 17. There has been a marked rise in paediatric IBD over recent decades.

IBS is often diagnosed on the basis of signs and symptoms, without a need for further investigations, but distinction from IBD on clinical grounds is often not possible. Blood tests that indicate the presence of inflammation (ESR and CRP) have been used as an aid to diagnosis, but may be abnormal because of other, non-gastrointestinal, conditions, and can be normal in people with IBD. Until recently, distinguishing between IBD and IBS has often required referral to specialist care for colonoscopy, an invasive and unpleasant investigation requiring sedation, usually carried out on a day case basis, at a cost of around £650 (including specialist referral and day case endoscopy). In younger patients, over 60% of colonoscopies have been normal, and in retrospect, not necessary.

Calprotectin is a protein released by the white blood cells involved in inflammation of the bowel. It is stable in faeces and can be measured by laboratory tests, and more recently by "point of care tests" (POCT). It indicates inflammation in the bowel, but cannot identify the cause of the inflammation.

The aim of this review is to examine the clinical effectiveness and cost-effectiveness of faecal calprotectin testing in distinguishing between "functional" disorders such as IBS, where sufferers will not come to serious harm, and "organic" disorders, such as IBD, that require referral to specialist care. In adults, the differentiation is most often between IBS and IBD. In children, there is a different range of conditions.

Perspectives on the use of calprotectin testing will vary with setting. General practitioners will see far more cases of IBS than IBD, and for them calprotectin testing offers evidence to rule out IBD. A negative calprotectin will imply IBS. So GPs will be looking for parameters such as sensitivity (for IBD) and negative predictive value, to provide a basis for a decision not to refer. These parameters provide measures of the risk of "false negatives" – patients with IBD, who should be referred but in whom the test is negative.

Gastroenterologists in adult clinics will be seeing a selected group of patients, referred by GPs, with a suspicion of IBD. Gastroenterologists will be looking for positive evidence of IBD in order to decide whether to proceed to further investigations, including colonoscopy and biopsy, and possibly also gastroscopy and other tests. They may find a positive predictive value or a positive diagnostics odds ratio more useful, because they will wish to avoid unnecessary invasive investigations in people who have IBS.

In effect, these are two sides of the same coin, based on the same data.

It should be noted of course that: diagnosis will be made on the whole clinical picture; that GPs are good at diagnosing IBS, for example on the basis of history, symptoms (gastrointestinal and other) and absence of weight loss; and that diagnosis would not be made on the basis of calprotectin results alone. However, GPs may find calprotectin useful to confirm a diagnosis based on clinical assessment.

The same general principles will apply to the different case mix seen in paediatric gastroenterology. For the GP, a high NPV from a normal calprotectin would lead to a decision not to refer to paediatric gastroenterology. The proportion with IBD is higher, but a normal or near normal calprotectin may contribute to a decision not to proceed to invasive procedures such as endoscopy, which requires either deep sedation or a general anaesthetic in children.

Methods

A systematic review and economic modelling were undertaken. A broad search strategy was run in several databases. Unpublished studies were also sought from the grey literature and personal

communication. Studies that provided sufficient data for calculation of sensitivity, specificity and other diagnostic outcomes were identified, and data was entered into Review Manager (Revman) version 5.2 for the generation of paired forest plots and receiver operating characteristic (ROC) curves. Further statistical analysis was performed in Stata 12 to produce likelihood ratios, area under the curve (AUC) and nomograms. Our intention was to examine the performance of calprotectin testing over a range of values, starting with the level recommended by the manufacturers, which is most often 50 µg/g. Where sufficient studies reported results at the same values, we aimed to pool data for each value. The quality of studies was assessed using the QUADAS 1 instrument. We sought studies in which the reference test was endoscopy with histology, but allowed a few with less invasive reference tests.

We also identified, appraised and summarised recent systematic reviews. Meta-analysis was performed in accordance with previously reported guidelines for meta-analyses of diagnostic tests using the Stata command Metandi. Pooled estimates for values among different diagnoses were obtained with 95% confidence intervals, assuming a bivariate model.

Results

Clinical effectiveness of calprotectin testing

The primary studies presented data for different groups of conditions, some providing a direct comparison of IBS and IBD, but others comparing a wider range of organic conditions.

Nearly all the evidence comes from studies in specialist care, with little data from primary care.

Seven studies gave results that compared IBS and IBD, at 8 cut-off levels, ranging from 8 to 150 µg/g, all in adults. Sensitivity was consistently high (usually 100% at levels under 50 µg/g; ranging from 83% to 100% at a cut-off of 50 µg/g), but specificity was more varied (51% to 100%), especially at lower levels of faecal calprotectin.

Two studies reported results for "organic bowel disorders" versus IBS. [redacted]

Eleven studies reported IBD versus non-IBD, with eight cut-off levels. They showed consistently high sensitivity at lower cut-offs, nearly all over 90%, with most at the 50 µg/g cut-off having sensitivities of 100%. Specificity was much more varied, ranging from 44% to 93% at a 50 µg/g cut-off. Most of these results were in paediatric groups. Most studies reported results at only one cut-off, but one reported five cut-offs and another four, both in paediatrics.

There were no studies based on a primary care population. Some were done in referrals from primary care, but GPs are selective in whom they refer.

The systematic reviews were of mixed quality and only one (unpublished but accepted for publication at time of review; high quality) was up to date.

Two reports by the York Health Economics Consortium were very useful. The first, from 2010, covered both clinical effectiveness and cost-effectiveness. It concluded that faecal calprotectin was a reliable marker of inflammation of the bowel, that high sensitivity was very important and that false positives were preferable to false negatives, that the cut-off should be 50 µg/g, and that in economic terms, calprotectin dominated (more correct diagnoses at less cost) blood tests such as ESR and CRP.

The second YHEC report was the final report (not yet published) from the pilots of implementation of calprotectin testing in Durham Dale and Northumberland. This provided useful data on the use of calprotectin testing in routine care and also on how it contributed to final diagnosis. One notable finding was that when GPs were sure a patient had IBS, they were usually right – 95% of such patients had normal calprotectin levels.

Choice of cut-off levels.
The commonest level for defining normality was 50 µg/g, as recommended by manufacturers, and in the 2010 YHEC report. If sensitivity was deemed of paramount importance (in order not to miss any cases of IBD), that level could be used. Some adults with IBS have raised calprotectin levels and would be "false positives", who might be referred for endoscopy as ? IBD. However there is some evidence that organic pathology is rare with levels under 100 µg/g, and clinical consensus is that if there are adults with IBD but calprotectin under 200 µg/g, they are likely to have low grade IBD and would come to no harm if simply monitored by repeated calprotectins, with referral if the level rose. In theory, a very sensitive approach might lead to people with IBS being false positives and being endoscoped, and a less sensitive approach might mean missing a few people with IBD, with more serious consequences. In practice, clinicians will apply clinical nous and observation, and that will reduce colonoscopies in false positives. Decisions will not be made purely on FC results.

In paediatric age groups, with a different spectrum of conditions, a cut-off of 50 µg/g gives almost 100% sensitivity but specificity varying from 44% to 94%. One study reported that a cut-off of 100 µg/g gave sensitivity of only 86%, specificity 91%. Another study recommended a level of 200 µg/g as being most useful in routine practice.

Cost effectiveness

NTAC pilot study

Results from a pilot implementation project in Durham and Dales CCG show that calprotectin testing could reduce costs of referral and investigation of patients under age 60 with chronic diarrhoea by over 60%, if all patients with negative tests are managed in primary care as IBS, with those with borderline and positive tests being referred to Gastroenterology.

This reduction is similar to the proportions of colonoscopies reported as normal from some other UK centres.

Review of previous studies

Previous economic analyses have typically concluded that faecal calprotectin testing is cost saving compared to the situation without it. Given test specificities and the assumed prevalences of IBD in the presenting population, the additional cost of the faecal calprotectin testing is more than offset by the reduction in the cost of unnecessary colonoscopies.

EAG assessment: primary care.

The EAG noted that without calprotectin testing, GP clinical assessment could be highly sensitive in referring IBD, and more so than calprotectin testing. However this was achieved at the cost of low specificity, with many "false positives" (people with IBS) referred to gastroenterology. GPs without faecal calprotectin testing would incorrectly identify 19.8% as false +ves requiring referral to colonoscopy. The rates of false +ves incorrectly referred to colonoscopy after CalDetect and ELISA testing would be much lower, 5.1% and 5.6% respectively.

Without calprotectin testing, many of the false positives would go on to colonoscopy, which has an extremely low risk of serious complications such as perforation. Such events are too rare to significantly affect costs, but they do have some QALY impact, as might the much more common minor adverse effects of colonoscopy.

Faecal calprotectin testing is estimated to result in cost savings. In theory, small QALY gains could accrue but these are too small to be significant, because of the low prevalence of IBD and the high sensitivities of all the tests, resulting in few false negatives with IBD. Some of the QALY difference arises from the very slightly lower mortality arising from a lower number of colonoscopies.

Sensitivity analyses around the base case suggest that faecal calprotectin testing results in patient gains and remains cost saving compared to GP assessment without faecal calprotectin testing, up to an IBD prevalence of 25%. At this point, due to ELISA testing having a less than perfect sensitivity, ELISA testing starts to result in very slight QALY losses compared to GP assessment without faecal calprotectin testing, though retains cost savings of around £63 per patient on average.

Secondary care

This applies most to the paediatric population. Despite the higher IBD prevalence in the secondary care population, the main test differences still lie in the number of unnecessary colonoscopies. Without faecal calprotectin testing all 52.1% of non-IBD patients receive a colonoscopy, compared to 13.5% for the ELISA with the 50µg/g cut-off and only 9.4% for ELISA with the 100µg/g cut-off.

The additional ELISA test costs are more than offset by the savings from reduced colonoscopies. Compared to referring all directly to colonoscopy, ELISA with the 50µg/g cut-off is estimated to save £205 on average, while ELISA with the 100µg/g cut-off is estimated to save £240. Trivial QALY gains of around 0.001 QALYs may occur with ELISA compared to direct referral to colonoscopy, these being slightly larger for ELISA with the 50µg/g cut-off due to its better sensitivity. But given the additional average £35 cost, the cost effectiveness estimate for ELISA with the 50µg/g cut-off compared to ELISA with the 100µg/g cut-off is £35,000 per QALY. As before for the primary care modelling, it should be stressed that the QALY differences between the faecal calprotectin tests are very small and they may better be considered as equivalent.

Research needs

There is a lack of studies in primary care populations.

Some people with IBS do have raised calprotectin levels. The reasons for that are not clear.

The remit of this review, as determined by NICE, was to evaluate calprotectin testing for differential diagnosis in newly-presenting patients. Calprotectin can also be used for monitoring disease activity and response to treatment, but consideration of that is outwith the scope of this review. However there is a need for further research in that use of calprotectin.

Conclusions

The NICE scope raised questions, abbreviated in italics below.

Is calprotectin testing a reliable way of differentiating inflammatory disease of the bowel from non-inflammatory ones?

Yes. Faecal calprotectin testing identifies patients with inflammation of the bowel, who need referral to specialist care. The majority of younger adult patients seen with lower abdominal symptoms in general practice have IBS, and the absence of inflammation as indicated by a negative calprotectin

test means that IBD is very unlikely. They can then be managed in primary care and spared further investigations.

What are the optimal cut-offs for use in primary and secondary care

The same cut-off should be used in primary and secondary care – 50µg/g for ELISA tests. This is based on ensuring high sensitivity, and not missing people with IBD. Some people assessed as positive by this cut-off will have borderline levels of 50 to 200µg/g, and may initially be monitored with repeat calprotectin testing, but some of this group will progress to definite IBD.

How do the rapid point-of-care tests compare to the laboratory tests?

There are few studies directly comparing tests, and on clinical effectiveness grounds, there is insufficient evidence to recommend one test over the others. The point of care tests can provide faster results. Costs vary amongst tests. None of the test kits are expensive but labour costs vary. The evidence base varies amongst tests. There are currently no grounds on either diagnostic reliability or cost-effectiveness considerations for preferring one test over another.

How will calprotectin testing perform in primary care?

Sensitivity and specificity will be as good in primary care, but the lower prevalence will increase the negative predictive value. The main benefit in primary care will be to confirm the clinical diagnosis by GPs of IBS. Making calprotectin testing available to general practitioners will greatly reduce the number of younger adults referred to specialist care, and the need for invasive investigations such as colonoscopy.

Impact in secondary care

In secondary care, the main benefit will be a marked reduction in colonoscopies that find no abnormalities. Calprotectin testing will considerably reduce the number of colonoscopies required. In various studies, over 60% of colonoscopies in this group of adult patients have been normal

Calprotectin testing will lead to considerable savings to the NHS, as well as the avoidance of an unpleasant invasive procedure in people whose symptoms are due to IBS.

Calprotectin testing can also reduce the need for colonoscopy in children who do not have IBD, and could reduce diagnostic delays in those who do. It could also reduce loss of work time for parents and loss of school time for children.

1. Introduction

1.1 The conditions

Chronic abdominal pain or discomfort, accompanied by diarrhoea or constipation, is common. The symptoms can be due to a number of different conditions, some more serious than others. The conditions include irritable bowel syndrome (IBS) and inflammatory bowel disease (IBD). The commonest forms of the latter are ulcerative colitis (UC) and Crohn's disease (CD, sometimes called regional ileitis, but that term is misleading because Crohn's can have a much wider distribution).

Lower bowel symptoms are very common in general practice. Most patients have IBS, a troublesome and painful condition that reduces the quality of life, but which does not have serious effects in terms of structural damage to the bowel. However some patients have IBD, which can lead to serious complications. In Crohn's disease, most patients will require surgery within 5 years. It is important to distinguish IBD from IBS so that patients with the former can be appropriately managed and monitored. IBD is characterised by inflammation of the bowel, which is not seen in most patients with IBS.

Unfortunately, the symptoms of IBD and IBS are often similar, and until recently, definitive diagnosis was often made only after invasive colonoscopy and perhaps other investigations. Faecal calprotectin testing identifies patients with inflammation of the bowel, who need referral to specialist care. The majority of patients have IBS, and the absence of inflammation as indicated by a negative calprotectin test means that IBD can be ruled out. They can then be managed in primary care and spared further investigations.

The most common symptoms of IBS include recurrent colicky abdominal pain or cramping felt in the lower abdomen and relieved by defecation. There may be abdominal distension (bloating) and altered bowel habit – episodes of diarrhoea and constipation. Features supporting a diagnosis of IBS include;

- symptoms >6 months
- bloating
- associated with other, non-GI problems
- symptoms worsened by stress.
- no weight loss

The Rome criteria subdivide IBS into diarrhoea predominant (IBS-D), constipation predominant (IBS-C), or mixed (IBS-M), with roughly a third in each group.

IBS is very common – perhaps 15% of the UK population, though many people who have it never consult their GPs about it. IBS-D is the commonest form.

It is commonest in young women, with an odds ratio in women: men of 1.7.[1] The IBS-C form is commoner in women than men. The underlying mechanism is not known. People who have it are constitutionally well and do not lose weight. It is a troublesome but not a serious condition, in the sense that it does not lead to serious adverse events. But it can be painful and disruptive of normal activities, and people with IBS have a reduced quality of life, reported to be reduced by 26% [2], and 30% if severe.[3] Quality of life is reduced because of disturbed work and sleep, and anxiety. It leads to 9 to 22 lost days of work per year.[4] Akehurst et al report that in the Trent Region, people with IBS had reduced quality of life compared to age, sex and socially matched controls, reflected in every dimension of both SF36 and EQ-5D, had more time off work, and imposed £123 more costs per year on the NHS.[5] The effect on quality of life depends on severity of symptoms, with those meeting the Rome II criteria faring worse than those meeting Rome I criteria.[6]

The British Society of Gastroenterology (BSG) commissioning report noted;
"While IBS is not a life-threatening condition, it is a major cause of ill-health and disability, disrupting social activity and work. The large number of patients affected, the need to screen out other diseases, and absenteeism and impairment in the workplace all constitute a major cost to the health service and society at large." [7]

The cause of IBS is not known in most people, but it sometimes follows an episode of infectious gastroenteritis ("food poisoning"). It is often associated with anxiety and depression, and bouts may be triggered by a period of stress.

An important point is that the symptoms of IBS, such as pain, can be quite severe, and may make sufferers think they have something more serious. As the British Society of Gastroenterology note;
"People fear that they may have cancer or that the doctor is missing something more serious. "Surely something as simple IBS would not make me feel so dreadful."
[8]

As we note later, this may affect referrals, if people seek reassurance by asking GPs to refer them to specialist care.

Conversely, many people with IBD do not consult their doctors until they have had symptoms for some time. A study from Germany reported that CD and UC patients waited for almost 8 months on average before consulting a physician.[9]

Coeliac disease is a disease of the small bowel, resulting from an immune reaction to the wheat gluten and similar proteins found in rye, barley, and, to a lesser extent, oats. Coeliac disease can be ruled out by testing for auto-antibodies at an early stage, so is not relevant to calprotectin testing. It could be classed as an inflammatory disease of the bowel, but the inflammatory cells are mainly lymphocytes, so calprotectin is not high (but can be modestly raised in children (D Wilson, personal communication.)

Ulcerative colitis is characterised by inflammation of the colon, sometimes intense, with bloody diarrhoea, but is often much milder. The cause is not known, but it appears that some people are more genetically susceptible than others.[10] Around 10% of people with UC have a first degree relative with the condition. The concordance in monozygotic twins is also around 10%.

Curiously, cigarette smoking may confer some protection, or reduce severity.[10] The risk is also moderately reduced in people who have had appendicitis and appendix removal, under the age of 20.

There may be an abnormal immune response to the microbacteria that normally live in the gut, known as commensals. UC is sometimes triggered by episodes of gastroenteritis caused by organisms such as Salmonella, Shigella and Campylobacter, but more by changes in the natural gut flora than direct effects of these organisms.

UC typically starts in the rectum and spreads upwards through the colon. The natural history is of relapse and remission. At first presentation, most patients have mild disease, and only 10% have severe disease. About half will continue to have mild disease or remission, but in about one fifth of patients, UC will be chronic and continuous, and more likely to become extensive, throughout the colon. By 10 years after onset, around 20% of patients will have required removal of the colon (colectomy).

The aim of treatment in active disease is to secure a remission, and then maintain that. Different drugs are used to induce, and then maintain, remission. There is an increased risk of colorectal cancer, so surveillance for that is part of care.

Crohn's disease can present in different ways, depending on which part of the intestinal tract is affected. Like UC, it is a relapsing and remitting inflammatory disease. However it can affect any part of the gastrointestinal tract – it is a much more extensive disease. Also like UC, there is a genetic susceptibility, with concordance in 35% of monozygotic twins.[11] The cause is unknown, but it appears to be commoner in those with a "westernised" lifestyle. Like UC, it may occur after infectious

gastroenteritis and is associated with disturbances in the usual gut flora. The histological features include those similar to tuberculosis but no mycobacteria have been shown to be responsible. There are around 60,000 people with CD in the UK, of whom 20-30% are aged under 20.[12] The incidence is highest in the age range 15 to 30 years. About 25% of cases have onsets under age 17.

The pattern of symptoms in children is different. A prospective survey was carried out in the UK and Ireland by the British Paediatric Surveillance Unit, the British Society of Gastroenterology Research Unit and the Paediatric Register of IBD. 739 cases under the age of 16 were reported, making it the largest such study. The commonest presenting symptoms of CD were abdominal pain, weight loss and diarrhoea, but 44% did not report diarrhoea, and only 25% reported the classical triad of abdominal pain, diarrhoea and weight loss. Other symptoms at presentation included lethargy and anorexia. Paediatric IBD (PIBD) is often more extensive at diagnosis than in adults.

The UK and Ireland survey found that delays in diagnosis of CD in children were common; 18% had a pre-diagnosis duration of symptoms of 1 to 3 years, and 9% of more than 3 years. Only 9% had isolated small bowel disease.

The delay in diagnosing PIBD has changed little over the last 20 years. What has changed is the incidence. Henderson et al reported a rise in Scotland of 76% from 1990-95 to 2003-2008, and a 5-fold increase over the last 40 years, especially in CD.[13] This rise may not apply to the same extent in the rest of the UK since there is a north-south gradient within Scotland[14], but internationally rates have been increasing.[15]

Symptoms of CD include diarrhoea, pain and blood or mucus in stools. Other presentations include anaemia due to disturbance of iron metabolism, and extra-intestinal disease such as arthritis, which may appear before any intestinal symptoms. Diagnosis is usually based on histology after biopsies taken during endoscopy. Differential diagnosis includes other causes of abdominal pain, such as IBS. Symptoms may be different in children, where growth retardation may be a feature that can precede bowel symptoms.[16]

The outlook in CD is worse than in UC. Only 10% have prolonged remission. Based on past experience, about 20% require hospital admission each year, and half will have required surgery within 10 years of diagnosis. This compares with the 10% of people with UC who will require a colectomy in the first 10 years.[17]
Newer drugs such as the “biological” agents (infliximab and adalimumab) may reduce admission rates and the need for surgery.[18,19]

There are three main serious intestinal complications of CD. The first is stricture (narrowing) of the bowel. This can lead to intestinal obstruction, and CD can present as an "acute abdomen" requiring surgery, sometimes mimicking appendicitis. The second is fistulas, which are abnormal connections between sections of bowel, or between bowel and bladder. The third is colorectal cancer, and surveillance is required.

In both UC and CD, some people have active disease but no symptoms. This has been noted following the introduction of colorectal cancer screening using faecal occult blood testing (FOBT). Positive screenees are referred for colonoscopy. Butcher et al reported that amongst 5350 such people who had colonoscopy, 66 were found to have unsuspected IBD (UC:CD 2:1) of whom about half had no symptoms. Some had quite extensive UC.[20]

Esch et al reported that some people with CD have no symptoms but are found by chance during investigations for other reasons.[21] However most developed symptoms over time (mean 3-4 years; range 2 months to 9 years) and a quarter required surgery. They concluded that initially silent CD requires similar monitoring to initially symptomatic CD.

The treatments and the aims of treatments have changed in recent times. Schoepfer et al comment that the aims have evolved from relieving symptoms towards mucosal healing.[22] They consider that this has been driven by the arrival of new medications such as the anti-tumour necrosis factor (anti-TNF) drugs that can induce and maintain mucosal healing. A New Zealand consensus conference concluded that early use of infliximab at induction, led to higher rates of mucosal healing.[23] Economic modelling by Ananthakrisnan et al suggests that treatment aimed at mucosal healing is cost-effective compared to aiming only at relief of symptoms, because over a 2-year follow-up period the mucosal healing group would have fewer hospital admissions and less surgery than the symptomatic-suppression group.[24] This results in a cost per QALY of around £33,000, based on straight conversion of $s to £s.

The arrival of more effective new drugs increases the importance of prompt diagnosis of CD, and it could be argued that they should be used earlier in the treatment pathway. However NICE TA 187 recommends use of the anti-TNF drugs, infliximab and adalimumab, only in people whose disease has not responded to conventional therapy with steroids or with immunosuppressive agents such as azathioprine and 6-mercaptopurine.[12]

The ratio of CD to UC varies between adults and children. In adults the ratio of CD:UC is 2:3[25], whereas in children the ratio is much higher CD:UC 2.3:1.[26]

Differential diagnosis

Some features of CD, UC, IBS and celiac disease are compared in Appendix 1. The key point is that distinguishing amongst them by purely clinical means – signs and symptoms – can be difficult. Ford and colleague carried out a systematic review of the usefulness of symptoms and symptom scores for diagnosing IBS.[27] They concluded that individual symptoms (lower abdominal pain, passage of mucus per rectum, feeling of incomplete evacuation, passage of looser stools at onset of abdominal pain, abdominal pain relieved by defecation and patient report abdominal bloating) have limited usefulness for diagnosing IBS. They also concluded that composite scores such as theManning and Kruis criteria had only modest accuracy, and noted that these scores were developed based on secondary care populations and might be less applicable to primary care patient mix. They also noted that around 40% of patients in the studies underlying the scores had some form of organic disease, suggesting an element of spectrum bias.

Jellema et al carried out a systematic review of the accuracy of symptom-based criteria for IBS (Manning, Kruis, Rome I and II and others).[28] They included 25 studies, but only three were carried out only on primary care patients. Jellema et al concluded that none of the criteria could reliably exclude organic disease.

However there is a school of thought that asserts that;
"a positive diagnosis of IBS should be reached using symptom-based clinical criteria, not after excluding organic disease by exhaustive investigation" [1]

This is echoed in the NICE scope,[29]
"*In the majority of cases the diagnosis of IBS can be made on the basis of clinical history alone."*

The systematic review done by the National Collaborating Centre for Nursing and Supportive Care for the NICE guideline group on IBS (page 101) quotes Jeong et all 1993;*"It is amazing to see the expensive, dangerous and extensive workups to which healthy patients are subjected by physicians searching for an organic cause in patients who obviously suffer from IBS."*[30]

The review lists many possibly investigations (pages 100-101) but these did not include calprotectin.

GPs in Durham Dale pilot were good at diagnosing IBS – if a GP thought a patient had IBS, the GP was right in 95% using a negative FC as confirmation of diagnosis. Note that this does mean that one in 20 patients had a diagnosis other than IBS, with raised calprotectin suggesting IBD.

It may be useful to consider new presentations separately. Many GPs will feel confident about the diagnosis in recurrent IBS, when they know the patient well and they have presented with similar

symptoms on previous occasions, perhaps after anxiety or stress. They may not feel a need to refer such patients. However with new presentations, there will be more diagnostic uncertainty, and the proportion referred to secondary care to exclude IBD may be higher. Calprotectin testing may be most useful in new presentations.

A survey of GPs from around Bristol found that most GPs were fairly confident (8 out of 10 where 10 was most confident) that they could diagnose IBS at the first visit and most did not investigate the under 45 year age range further. However since only a small proportion were referred for specialist investigation, there may have been some false negatives with IBD [31]

However, many patients are referred to gastroenterology, for definite diagnosis, which is usually/often based on endoscopy and histology of biopsies. Some studies report that some patients with IBS are very anxious, and require the reassurance of a hospital "check-up". In one small study (54 patients) from Cardiff, the main reason for referral was diagnostic uncertainty (37 of 54) but the second reason was for "confirmation of IBS" (17/54).[32]

In various studies, the proportion of patients referred for further investigation in whom abnormal findings are reported on colonoscopy, is low. Kok et al noted reports that only 22% to 37% had organic bowel disease.[33]

The ability of GPs to correctly identify as IBS a considerable proportion of people with lower abdominal symptoms, has implications for the spectrum of patients in whom calprotectin testing might be used. IBS is very common, and one estimate is that 90% of patients seen in general practice with chronic lower abdominal symptoms have IBS. This high prevalence of IBS in general practice groups has led to concern that results from studies carried out in secondary care may not be applicable to patients seen in primary care. A much higher proportion of patients in secondary care studies may have IBD. However, if GPs are referring only selected patients to specialist clinics, the prevalence of IBD amongst referrals will be higher, with the spectrum of referred patients more similar to that in the studies from secondary care.

Endoscopy can be: 1) colonoscopy, involving inspection of the whole colon, 2) sigmoidoscopy, inspection of only the distal part of the bowel (the sigmoid colon), or 3) gastroscopy, visualising oesophagus, stomach and upper part of the small bowel. There are some sections of the small bowel that cannot currently be reached by widely available forms of endoscopy. In those situations, options include capsule camera endoscopy (the "camera pill"), and imaging methods including ultrasound and MRI.

Long delays in diagnosing IBD have been reported. Burgmann et al from Manitoba reported that 42% of a group of people with known IBD, had had gastrointestinal symptoms for more than 3 years before the diagnosis of IBD, with some having symptoms for as long as 11 years before IBD diagnosis.[34] Delays were much commoner in older age groups, with an incorrect diagnosis in around half the over 64s compared to only around 10% in younger adults.

1.2 NICE clinical guideline 61 (IBS in adults)

The National Institute for Health and Clinical Excellence (NICE) clinical guideline 61 makes recommendations for adults with irritable bowel syndrome (IBS).[35] The guideline recommends that patients with IBS should to be encouraged to manage their symptoms by themselves initially, and be given information on general lifestyle, physical activity, diet and symptom-targeted medication.

The advice on diet should be tailored according to the patient's symptom (diarrhoea, constipation). If diarrhoea is the predominant symptom, then patients should be advised to limit intake of high-fibre food, limit the consumption of fresh fruit and avoid eating insoluble fibre. Patients should also avoid consumption of sorbitol (an artificial sweetener) found in sugar-free sweets and drinks.

If the predominant symptom is constipation, then patients should be advised not to consume starch that resists digestion in the small intestine and reaches the colon intact. If patients need high dietary fibre then they should take soluble fibre such as ispaghula powder or foods such as oats that are rich in soluble fibre.

Some of the advice that relates to all types of IBS includes: having frequent meals and eating slowly; do not skip meals or having long gaps between meals; drinking at least 8 cups of fluid per day especially water; restricting tea and coffee to three cups per day; avoiding insoluble fibre.

If patients continue to have symptoms and severity increases, then pharmacological intervention is recommended, but no length of time before this is specified.

IBS-diarrhoea

Pharmacological intervention

First line treatment: antispasmodic agents should be taken as and when required, alongside dietary advice. Loperamide is the first choice agent.

Second line treatment:

Tricyclic antidepressants (TCAs) started at a low dose taken at night. If TCAs are ineffective, then selective serotonin reuptake inhibitors (SSRIs) can be tried. After prescribing TCAs or SSRIs, patients should be followed up after 4 weeks and then 6 to 12 monthly intervals thereafter.

Psychological interventions

If patients do not respond after 12 months of pharmacological therapy, they may be referred for psychological interventions such as cognitive behavioural therapy (CBT).

IBS-constipation

Pharmacological intervention

First line treatment:

Ispaghula powder (laxative). Further management in patients with IBS-constipation is similar to those with IBS-diarrhoea.

The reason for including the above summary is because it shows that IBS may be treated in a stepwise way. Each step may take time to be tried, and many patients will not respond to the first or later therapies. The importance of this is because a patient with IBD, misdiagnosed as IBS, may go through a time-consuming series of treatments for IBS, before clinical suspicion leads to referral to gastroenterology or paediatrics. IBS can cause considerable pain and discomfort, sometimes more than IBD.

1.3 Calprotectin

Calprotectin is a protein found in some cells, most notably the group of white blood cells called neutrophils. It binds to calcium, and is then a stable compound not broken down in the intestines.

In people with bowel conditions that cause inflammation, the increased number of neutrophils in the bowel leads to an increase in faecal calprotectin. It can therefore be used as an indication of inflammation. There are now tests to detect or measure the level of calprotectin in faeces. It appears stable in faeces for at least 7 days (though not all agree). It is also reproducible from day to day in individuals. Naismith et al obtained stool samples on three consecutive days from 143 patients with CD and found low day to day variation.[36] They concluded that clinical decisions could be made on a single calprotectin result.

Moum et al reported considerable variability in FC levels in patients with CD, in samples taken on two consecutive days.[37] However the variability was seen mainly at high levels, with little in the borderline region of 50-200 mg/l (normal is under 50).

There can be false positives from the taking of non-steroidal anti-inflammatory drugs (NSAIDs), but these can be avoided by asking patients to stop taking the drugs before calprotectin testing.
In a Finnish study that compared medication use amongst people with IBD, and the general population, people with IBD had almost a fourfold increase in use of PPIs (OR 3.9) and a slight increase in the use of NSAIDs (OR 1.17).[38] However not all studies have reported increases with NSAIDs. In a study amongst those with borderline calprotectin levels (>50 but <150), Demir et al found no significant difference with NSIAD use.[39] Conversely, Turvill reported that 14% of people referred from primary care with intestinal symptoms, and who had raised calprotectin, had a final diagnosis of NSAID enteropathy.[40]

There can also be false positives after chest infections (because of the white blood cells in swallowed sputum) and after bleeding into the bowel.

The proposed role of faecal calprotectin (FC) testing in this appraisal is for supporting differential diagnosis in people with lower gastrointestinal symptoms (pain, bloating, diarrhoea, change in bowel habit). The aim is to distinguish between those with inflammatory conditions and those with no inflammation. Many of those with inflammation will have IBD, but others may have cancer or other conditions. Most of those with no inflammation will have IBS.

Knowledge of the presence or absence of inflammation will affect the decision on referral for further investigation. The absence of inflammation may lead to a presumption of IBS, to be managed in primary care. The presence of inflammation would be likely to trigger referral to gastroenterology for further investigation, likely to include endoscopy.

Hence there could be two benefits. Those with IBS would not be referred and might therefore escape further investigations especially colonoscopy. Those with inflammation might be referred more promptly and receive appropriate treatment earlier.
FC could be part of a pre-referral work-up in general practice, such as outlined in Figure 1. In the second box, TTG refers to testing for coeliac disease. The term “red flag” is used to refer to symptoms or signs that might be due to cancer, including anaemia, rectal bleeding, unexplained weight loss, abdominal masses, and change in bowel habit in patients over 60 years of age. A family history of bowel cancer might also be a red flag item.

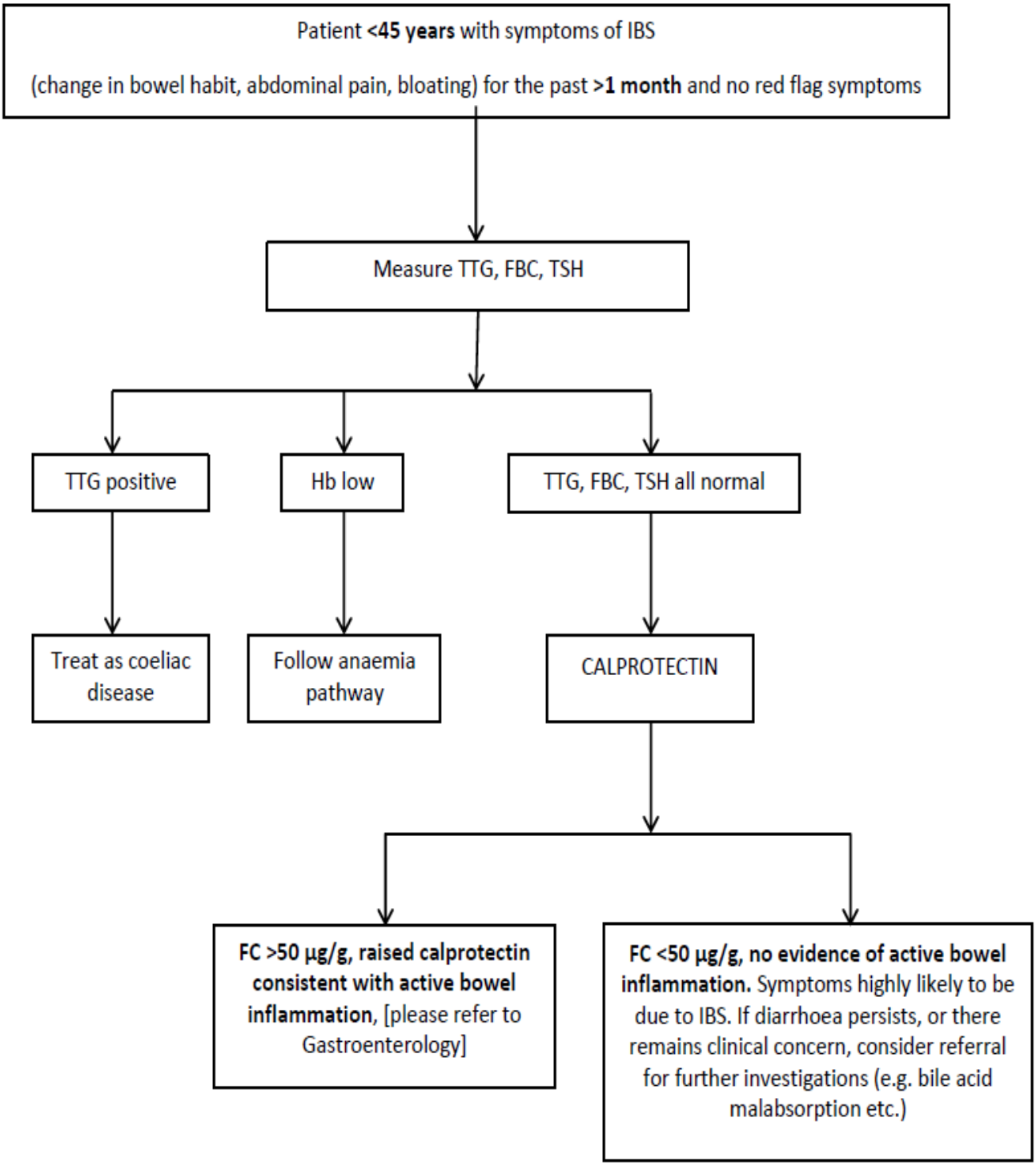


Figure 1. Possible pathways in patients with symptoms of IBS

The age cut-off of 45 is somewhat arbitrary, but was used in the BSG guidelines for diarrhoea in 2002.

The first stage involves excluding patients with "red flag" signs or symptoms. These could be indicative of cancer and are indications for rapid referral. However, many people with proven IBS also have red flags. Whitehead et al report data from the Puget Sound Health cooperative.[41]

Table 1. Red flag indicators in IBS and cancer

	IBS	GI cancer
Blood in stools	15%	14%
Unintended weight loss	21%	56%
Onset of symptoms after age 50	32%	67%
Family history of cancer	20% (unclear, but presumed colon cancer)	39% (colon cancer)

Rectal bleeding may be due to haemorrhoids ("piles") which are common (around 20%) in patients with IBS, especially those with IBS-C.

The next stage involves blood tests, one of which is TTG (tissue transglutaminase) a test for coeliac disease. This means that coeliac disease can be confirmed or ruled out at this stage. At present this stage also involves measurement of ESR (erythrocyte sedimentation rate) and CRP (C-reactive protein) which are markers for inflammation. However these do not localise the inflammation to the bowel, whereas FC does.

One issue to be considered is whether ESR and CRP should be done at the same time as TTG and FBC, on the grounds that they are cheaper, and can done at the first visit. If negative ESR and CRP could rule out inflammatory conditions of the bowel, a presumptive diagnosis of IBS could be given, and referral for further investigations would not be made at this stage. However a number of studies have reported that CRP and ESR have poor sensitivity and/or specificity[42,43] Tomkins unpublished manuscript) meaning that they are negative in many people with active CD. The report by the York Centre for Health Economics (YHEC) for the Centre for Evidence-based Purchasing (CEP) concluded that faecal calprotectin testing dominated (i.e. was both more effective and less costly than) ESR and CRP.[44] More recently, Mascialino et al from one of the manufacturers of calprotectin tests, Thermo Fisher Scientific (Phadia AB, Uppsala, Sweden) also concluded that faecal calprotectin testing dominated ESR and CRP, after taking into account all costs, in primary and secondary care, including

reductions in endoscopies.[45] Their estimate for the UK was that FC saved at least £100 per patient investigated, compared to ESR and CRP.

Not all forms of inflammatory colitis will increase calprotectin. There are some rarer forms, such as lymphocytic colitis. Lymphocytes are another form of white blood cells, but unlike neutrophils, they do not release calprotectin.

1.4 Decision problem

The aim of this review is to examine the clinical effectiveness and cost-effectiveness of faecal calprotectin testing in distinguishing between "functional" disorders such as IBS, where sufferers will not come to serious harm, and "organic" disorders, such as IBD that require referral to specialist care. In adults, the differentiation is most often between IBS and IBD. In children, there is a different range of conditions.

If calprotectin is a reliable way of detecting inflammation of the bowel, or its absence, then those patients in whom the test shows normal levels could be spared referral to specialist care and the often invasive and unpleasant investigations, such as colonoscopy, that may follow.

1.4.1 Population

The population is patients with lower gastrointestinal symptoms that are chronic, defined as persisting for at least 6-8 weeks. The upper age limit is 60 years, as per the NICE scope.[29] Symptoms in adults include abdominal pain or discomfort, bloating or change in bowel habit. Some will be newly presenting in primary care; others may already have been referred to specialist care.

Children (under 17) are a separate group with a different mix of conditions.

The main focus would ideally be in primary care, because that is where people with lower bowel symptoms first present. Faecal calprotectin testing has not been widely available in, or to, primary care, and hence much of the differential diagnosis has been done in hospital clinics.
This could potentially give rise to problems reflecting selection for referral. For example, there may be three groups of people with IBS;

- those who do not seek help or advice from GPs, but self-treat as required, with over-the-counter medications such as laxatives and analgesics. "Self-managed"
- Those who do present to their GPs, but whose symptoms are not considered such as to require referral. "GP-managed"

- Those referred by GPS to specialist clinics – the referred group. At present it is estimated that only about 25% of patients are referred to secondary care. However, since in the past, referral was often followed by colonoscopy, the threshold for testing with calprotectin may be rather lower than the threshold for referral, and many more than 25% may be tested with calprotectin. (Later, we assume 50%).

Evaluation of tests only on the referred group could, at least in theory, cause spectrum bias problems if the prevalence of IBS was less, and that of IBD higher, since parameters influenced by prevalence might differ from the GP-managed group. This could be important if FC testing was recommended and made more widely available. However as noted above, GPs are highly selective in whom they refer.

Testing will be used mainly for the diarrhoea form of IBS (IBS-D) and not the constipation form (IBS-C).

1.4.2 Intervention

Faecal calprotectin tests. These are of two types;

- Laboratory testing using ELISA methods
- Point of care testing (POCT), which can be used in primary care or secondary care.

Laboratory methods are quantitative. POCT tests may be quantitative or semi-quantitative.
The POC tests can give faster results, within about 30 minutes (which may be quite slow in the context of the pressure of work in general practice.) Extraction of the faecal sample is always manual so some time costs are irreducible.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

Table 2. List of faecal calprotectin tests

Name of Test	Type of test	Manufacturer	
Phical ELISA kit	ELISA	Calpro (Oslo, Norway)	Quantitative ELISA using polyclonal rabbit antibody. Recommended cut-off 50 µg/g
PhiCal Calprotectin ELISA kit	ELISA	Immunodiagnostik AG (Benheim, Germany)	Quantitative ELISA, using two monoclonal antibodies. Recommended cut-off 50 mg/kg, and can also be used in children aged 4 to 17 years. The manufacturer recommends that laboratories establish their own reference range.
EK-CAL	ELISA	Buhlmann Laboratories (Switzerland)	Monoclonal antibody. Two ranges with low range used for FC levels up to 600µg/g (range 10-600µg/g). The cut-off level is 50µg/g for both adults and children aged between 4 and 17 years.
Calprest	ELISA	Eurospital Spa (Trieste, Italy)	The cut-off level is 50 mg/kg. The manufacturer suggests retesting after a short period of time in patients with FC levels between 50 and 100 mg/kg
CALPRO Calprotectin ELISA test (ALP)	ELISA	CALPRO AS (Lysaker, Norway)	quantitative method Normal range up to 50
Quantum Blue	POCT	Buhlmann Laboratories	Two types rapid tests i) the lower range (30 to 300 µg/g) and high range(100 to 1800 µg/g). cut-off value of this test is 50 µg/g. The manufacturer recommends re-testing samples if results are between 30 and 70 µg/g. This zone is regarded as 'grey zone' and the values corresponds to the 2.5^{th} -97.5^{th} percentile of imprecision around the cut-off of 50 µg/g.
Prevent ID Caldetect	POCT	Preventis, GmbH (Bensheim, Germany)	Semiquantitative rapid test, with levels indicated by 3 bands:<15; 15-60; >60.
Prevista	POCT	GmbH & Co KG (Munich, Germany)	Semiquantitative immunochromatographic rapid test; recommended cut-off of 50 µg/g
EliA platform	EliA	Thermo Fisher (previously manufactured by Phadia)	EliA platform is a fully automated test, said by the manufacturer to reduce technician workload, time and cost.

1.4.3 Comparators

The main comparator is clinical assessment, which can be supplemented by ESR and CRP, which can indicate inflammation, but not localise it. There are two options for ESR and CRP testing;

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

1. If GPs have access to faecal calprotectin testing, they could use that in people with suspected IBS. So FC would replace ESR and CRP testing.
2. If normal ESR and CRP could exclude inflammation of the bowel, they might be used as part of the initial work-up. However the evidence suggests that normal CRP results can occur in the presence of active inflammation.

The limitations of ESR and CRP are that;

- Negative tests do not exclude IBD, so if symptoms persist, patients would still require further investigation
- Positive tests might be due to other, non-GI inflammations, so further investigations would be needed to localise the inflammation.

In one survey carried out in 2010, 89% of gastroenterologists considered calprotectin to be more accurate than CRP and ESR for distinguishing between IBS and IBD.[22] A review by Burri and Beglinger noted that ESR and CRP had low sensitivity.[46]
As noted earlier, CRP and ESR have poor sensitivity for IBD.
There therefore seems little point in doing these tests even if calprotectin was not available. As noted previously, the YHEC report noted that CRP and ESR were economically dominated by calprotectin. These tests are therefore not examined further.

1.4.4 Outcomes

Depending on data availability, these may include;

- Referral rates
- Numbers of colonoscopies with/without FC testing
- Proportion of colonoscopies with no abnormal findings
- Duration from onset of symptoms to definite diagnosis of IBD – late diagnosis of CD
- Cost
- Adverse events such as complications of colonoscopy
- Quality of life and hence QALYs

1.4.5 Modelling approach

A set of possible pathways is shown in Figure 2;

- No FC testing available. Clinical assessment and simple tests in primary care followed by decision on referral or symptomatic treatment/ therapeutic trial in those thought to have IBS
- Laboratory testing available to GPs. The laboratory just reports the results.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

- "Laboratory plus" where the GP provides clinical details along with test request and gastroenterologist or clinical biochemist provides commentary and advice
- POCT available in primary care. If it is negative, the GP manages the patient on a presumptive diagnosis of IBS. If the result is positive, the GP can refer to Gastroenterology for further investigation. If indeterminate, the GP can either repeat test or refer.

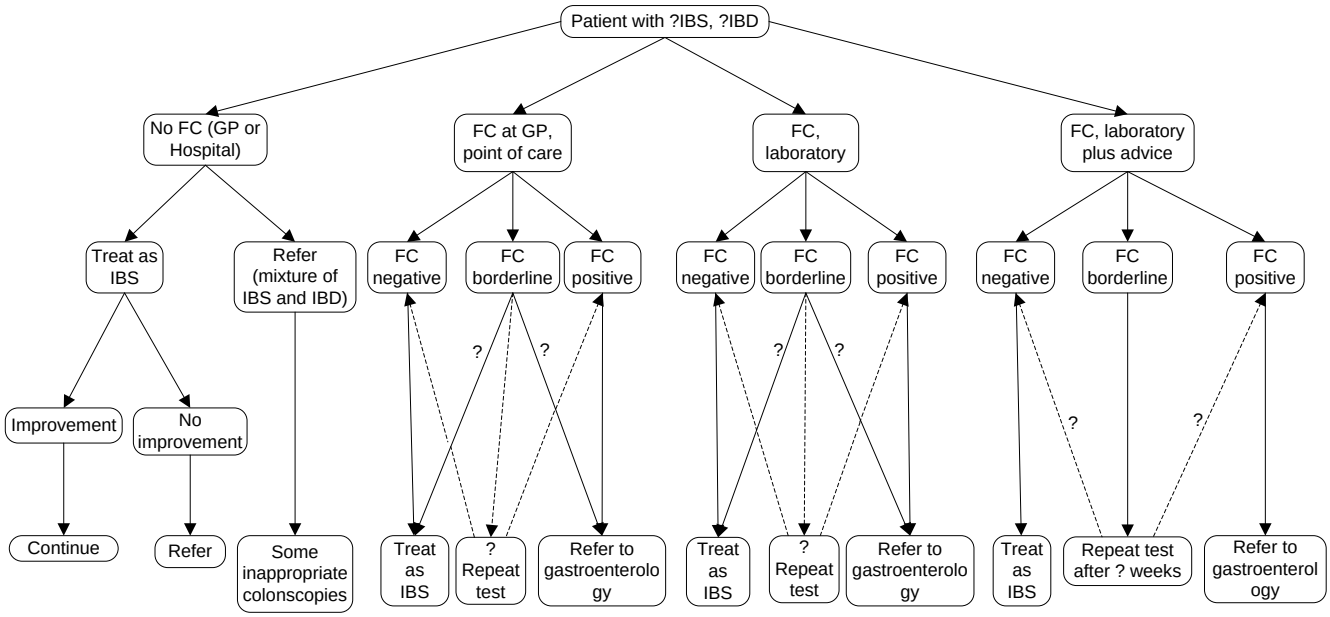


Figure 2. Service options

1.5 Methods

The inclusion criteria were studies comparing faecal calprotectin as a guide to inflammation of the lower intestine, ideally with histology as the reference test, in newly presenting patients. Exclusion criteria included studies of faecal calprotectin for monitoring activity of IBD or response to treatment in people with known IBD.
We also identified, appraised and summarised recent systematic reviews.

The databases searched for diagnostic studies included the databases Medline, Embase, Cochrane Library and Web of Science from their inception up to March 2013. Also, additional sources of grey literature were searched, the reference lists of relevant articles checked, and experts contacted for unpublished data. Full details of the search strategy are shown in Appendix 2.

The selection was done in three stages, based on fulfilling each of following criteria.

a) Were the patients newly diagnosed?

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

b) Was an acceptable reference standard used?
c) Were the appropriate outcomes reported; i.e. were sensitivity and specificity data reported or was it possible to derive a 2x2 table to determine them?

The hierarchy of evidence based on reference tests was;

1. Gold standard - endoscopy (usually colonoscopy) and histology.
2. Endoscopy and results by disease but no mention of histology – biopsies presumed to have been done.
3. Endoscopy with report that no biopsies done. Camera endoscopy included here
4. No endoscopy but diagnosis by imaging methods, for example thickened gut wall on CT.
5. Clinical follow-up for 6 months

Studies were grouped according to the conditions being compared, with most weight being given to;

- Studies comparing IBS with IBD
- Studies comparing IBD with all non-IBD conditions

Data were extracted from the included studies for 2x2 tables, with FC as screening test and bowel histology as the reference test. If studies fulfilled the other inclusion criteria, but data for 2x2 tables was not available, we reported what data were available, such as calprotectin ranges, medians, and IQR ranges, to compare groups with different conditions.

In papers where the numbers of true and false positives and negatives were not reported, but data on sensitivity and specificity and the total numbers of people with and without disease was reported, the data for the 2x2 table were calculated using the Calculator function in RevMan.

Data on five covariates, including FC cut-off level, make of test, age (adult or paediatric), setting (primary or secondary care), and type of test (ELISA or POCT) were extracted for each study and entered into RevMan.

All calprotectin levels were reported in ug/ml (or equivalent) apart from Tibble 2002[43] which used a non-commercial in-house ELISA, with levels were reported in mg/L. On the basis of data in previous systematic reviews, results were converted to µg/ml by multiplying by a factor of five.[47,48]

Statistical methods

Review Manager version 5.2 was used for data entry and analysis to generate forest plots. MedCalc version 12.3.0 for producing statistical data based on the 2x2 tables, including PLR, NLR, PPV, NPV and disease prevalence.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

Studies that provided sufficient data for calculation of sensitivity, specificity and other diagnostic outcomes were identified, and data was entered into Review Manager (Revman) version 5.2 for the generation of paired forest plots and receiver operating characteristic (ROC) curves. Further statistical analysis was performed in Stata 12 (StataCorp, 4905 Lakeway Drive, College Station, Texas 77845 USA) to produce likelihood ratios, area under the curve (AUC) and nomograms. Our intention was to examine the performance of calprotectin testing over a range of values, starting with the level recommended by the manufacturers, which is most often 50 µg/g. Where sufficient studies reported results at the same values, we aimed to pool data for each value.

Meta-analysis was performed in accordance with previously reported guidelines for meta-analyses of diagnostic tests using the Stata command Metandi.[49,50] Pooled estimates for values among different diagnoses were obtained with 95% confidence intervals, assuming a Bivariate model.

Meta-analysis was performed in accordance with previously reported guidelines for meta-analyses of diagnostic tests using the Stata command Metandi.[51]. Pooled estimates for values among different diagnoses were obtained with 95% confidence intervals, assuming a Bivariate model. If there were sufficient studies, we planned to pool data at the same cut-off levels from ELISA and POCT tests separately, and compare them. However only ELISA tests were pooled.

Quality assessment

Quality assessment of studies was done using items adapted from the QUADAS I tool (in protocol as approved by NICE), with questions formulated as follow.[52]

Quality assessment items used

1. Was the spectrum of patients representative of the patients who will receive FC testing in practice?
2. Is the reference standard likely to classify the target condition correctly? The reference standard for confirmation of bowel inflammation was histology of biopsies obtained at endoscopy.
3. Is the time period between FC measurement and obtaining tissue for histology short enough to be reasonably sure that the target condition did not change between the two tests? We regarded an acceptable delay between tests as being three months or less.
4. Did the whole group receive verification by histology? If not, were results for those who did receive histology reported separately?
5. Did patients receive endoscopy and histology irrespective of the FC result? (differential verification avoided)
6. Disease stage. Were patients newly presenting with symptoms? Some studies had mixed groups

of newly presenting and patients already known to have IBD, and we allowed up to 20% of non-new patients. Studies in patients with > 20% confirmed IBD, whether active or in remission, were excluded. Some studies clearly stated that patients were newly-presenting. In others (Damms 2008[53], Garcia 2006[54], Li 2006[55], Licata 2012[56], Shitirt 2007[57]), less detail was given, and we inferred that they were newly-presenting from terms such as "referred for investigation of chronic diarrhoea". So possible answers were yes, or probably. Ideally, we would have contacted authors, or excluded studies in which new presentation was not clear. However the number of studies was too low to adopt that approach.

7. Were histology or endoscopy results interpreted without knowledge of the FC results? (index test results blinded)
8. Were the FC results interpreted without knowledge of the results of histology? (reference standard results blinded)
9. Were the same clinical data available when test results were interpreted as would be available when the test is used in practice? (relevant clinical information)
10. Were intermediate FC results reported? (uninterpretable results reported)
11. Were withdrawals from the study explained? (withdrawals explained)

Question 6 replaced the usual QUADAS question on whether the reference test was independent of the index test, since histology, our preferred reference test, is clearly independent of calprotectin, so the usual question 6 would not help discriminate.

The term "quality assessment" is preferred to the more traditional "risk of bias" term because the latter, as used in systematic reviews such as Cochrane ones, is more associated with assessing internal validity of RCTs. We need to assess external validity through items such a spectrum bias.

All data extractions and quality assessments were done by one author and checked by another.

1.6 The NTAC pilot studies

The NHS Technology Adoption Centre (NTAC) has sponsored two pilot studies of the implementation of faecal calprotectin testing. Details as given in the NICE scope are as follow;

> *West Northumberland CCG is using a fully quantitative test (Quantum Blue) with samples being analysed in the laboratory. It is technically possible to use this equipment as a point of care test in primary care, although it is thought unlikely that this would ever be economical in practice.*
>
> *Durham Dales CCG is using a semi-quantitative point of care test (CalDetect, version 1), with the analysis being carried out in the GP Practice.*

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

In both cases there is a high cut-off value above which the patient should be referred to secondary care, and a low cut-off value below which there is a low probability of organic disease. Between the high and low cut-off values there is an intermediate range, in which case the patient should be retested. Due to differences in the assays used there is a difference in the cut-off values used in the project sites.

A cost-consequence analysis will be performed (by NTAC).

Data from the pilots are expected to help offset the current lack of data from primary care.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

2. Results of clinical effectiveness review.

The database searches retrieved 1273 references and 35 came from additional searches; there were 725 references remaining after de-duplication. The flow chart is provided in Appendix 2.
All of the 83 full text articles assessed for eligibility were assessed independently by three authors and any differences were resolved by discussion.

In the interests of brevity, studies in this section will usually be referred by name of first author and year.

Full details of the baseline characteristics of all the included studies are given in Appendix 6.

2.1 Some issues

A number of issues, listed below, arose in this review.

2.1.1 Reference standard

We took histology after endoscopy to be the definitive reference standard. Some studies used other reference tests. For example, the authors of one study in a paediatric group, quite reasonably, did not consider it justifiable to endoscope children with normal calprotectin levels. Instead, they used a six-month period of observation. In another study in adults, only those with high calprotectin levels had endoscopy. Those with normal levels were managed in primary care.

Note that not all CD can be reached by endoscopy. About 30% of CD in adults is ileal alone, and 50-60% ilio-colonic. But about 20% is in proximal or mid-ileum, so not accessible by standard colonoscopy of gastroscopy. Small bowel enteroscopy is complex, expensive and available in only a few centres in the UK. So options include video capsule camera or MRI.

MRI of the small bowel (especially after preparation with enteroclysis) is sensitive and has been shown to correlate with FC levels. Zippi et al reported a good correlation between MRI changes (such as wall inflammation and thickening of bowel) and FC levels.[58]

Ultrasound has been used in several studies. Aomatsu et al used ultrasound to detect CD in the small bowel in children, using > 3mm thickening of the small bowel as indicating active CD.[59] Calprotectin levels were much higher (mean 738) in children in clinical remission but with active lesions on ultrasound, than in children in clinical remission but without activity on ultrasound (mean 18). In this study, ultrasound was used as a reference standard for calprotectin testing, but the reverse can apply.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

Canani et al also used ultrasound in children but found some overlap between CD and non-IBD cases.[60] However they described transabdominal ultrasound of bowel wall thickness to be a useful and non-invasive method in confirming IBD, especially if inflammation was localised to the ileum.

Ultrasound is useful more as screening tool, since it is not sensitive enough to assess location of IBD. Small bowel MRI with contrast follow-through is standard in children. Wireless capsule endoscopy is also used.

Tomas et al considered that calprotectin showed good correlation with scintigraphy with radio-labelled leukocytes, which they considered was the gold standard for measuring inflammation in the bowel, though undesirable in children because of the radiation and the need for anaesthesia, and not used.[61]

2.1.2 Patient groups in studies.

The proposed value of calprotectin testing in primary care is to help GPs make decisions on likely diagnosis, in order to decide whom to refer to specialist care for further investigation. Patients with "red flag" signs or symptoms are referred, so are excluded from the calprotectin pathway. So the value of calprotectin is to guide decisions on whether to refer or not. A low calprotectin level indicates absence of inflammation, suggesting that IBS is the likeliest cause of the symptoms. A high level in someone with chronic symptoms suggests inflammatory bowel disease, CD or UC. (FC can be raised in acute bacterial gastroenteritis but that usually resolves rapidly.)

Many studies compared "non-organic" conditions (principally IBS in adults) with any organic condition. However some organic conditions are not obviously inflammatory, so studies where the organic group included a mixture of conditions could make calprotectin testing look less useful. Calprotectin will therefore appear most impressive in studies that include only IBD and IBS.

The table below shows two things. Firstly, the overlap in calprotectin levels between some organic conditions and IBS. Hence comparing only "all organic" and non-organic will make calprotectin testing seem less valuable. Secondly, that calprotectin levels are raised in colorectal cancer, and to a lesser extent in people with larger adenomas. Adenomas are not usually regarded as being inflamed, in the sense of being infiltrated with white blood cells.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

Table 3. data from Kok et al [33] on FC levels (µg/g) and adenomas

Calprotectin test	Adenocarcinoma	Adenomas ≤1cm	Adenomas >1cm	IBS
Quantum Blue	Median 215 IQR 105-300	Median 42 IQR 30-105	Median 111 IQR 30-264	Median 40 IQR 30-69
EK-CAL	Median 274 IQR 94-442	Median 60 IQR 24-108	Median 89 IQR 34-217	Median 49 IQR 21-99

2.1.3 Cut-offs for calprotectin

One problem with the evidence is that many studies used only the manufacturer's recommended cut-offs. This presents a problem when it comes to assessing optimum thresholds – there is little evidence for levels other than 50 µg/g. We are grateful to Professor K.D.Bardhan, Dr P Basumani and Dr A Banerjee for providing unpublished data from Rotherham on different cut-offs.

There is debate about the minimum number of studies that should be used for pooling data on different cut-offs with four being regarded as the minimum.[51] We have therefore not pooled studies if there were fewer than four at the cut-off in question, but have relied on diagnostic odd ratios as the summary statistic when there were fewer than four studies.

2.1.4 Spectrum

NICE is interested in the use of FC testing in primary care, but nearly all studies come from secondary care. The secondary care studies will have a different mix of patients from those seen in primary care. (See prevalence data in later tables.) The sensitivity and specificity of testing will be the same, but the different prevalence will give different predictive values.

This may be a particular problem in comparing different tests, such as point of care and laboratory tests. These may appear comparable in secondary care populations, but if the calprotectin levels are much higher in those selected populations, the comparability results may not be generalizable to populations with lower calprotectin levels.

However the only published studies comparing tests come from secondary care, and so we have had to use those.

Another issue about spectrum of patients arises from another selection effect.The pilot studies of calprotectin use in primary care from the northeast of England have shown that GPs are good at diagnosing IBS. In the Durham Dale pilot, 95% of those predicted by GPs to have IBS, had it. The GPs were also good at predicting IBD – 88% of patients who had high calprotectin levels, had been predicted by their GPs to have IBD. So GPs may confidently diagnose IBS on clinical grounds in many patients, which implies that those who will have calprotectin testing may be a selected group.

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

They may be more akin to those seen in secondary care – making the results from the secondary care studies more generalizable. A review by von Roon et al, described in detail below, concluded that individual GI symptoms could not reliably distinguish between IBS and IBD, but GPs may use non-GI symptoms or signs, and "clinical nous", to diagnose people with IBS.[48] A recent BMJ review of IBS concluded that it could be diagnosed on clinical grounds;[1]

"*The diagnosis should be reached using symptom-based clinical criteria, rather than excluding underlying organic disease by exhaustive investigation"*

2.1.5 Choice of measure

As noted by Harbord and Whiting, there is no single measure of diagnostic accuracy.[49] They recommend that the measures most often used are sensitivity and specificity, with the trade-off between these being illustrated graphically.

In the sections that follow, we report;

- Brief details of the included studies
- QUADAS quality assessment
- Results
- Sensitivity and specificity in paired forest plots, for all included studies
- For one study with a range of cut-off points we produce its own forest plot and ROC curve
- ROC curves with pooled sensitivity and specificity, and AUC
- Forest plots for the studies included in the ROC curve
- Fagan's nomograms with likelihood ratios
- Tables of diagnostic odds ratios for different cut-offs, pooled where appropriate

2.2 Previous reviews

Five recent systematic reviews were quality assessed and summarised. (see

Table 5,Table 6, and Table 7)

The 2010 review by the York Health Economics Consortium for the Centre for Evidence-based Purchasing provides a good starting point, since it was done to inform the debate about the value of calprotectin in identifying people whose symptoms were due to IBS, and who therefore did not need expensive and invasive investigations such as colonoscopy.[62]

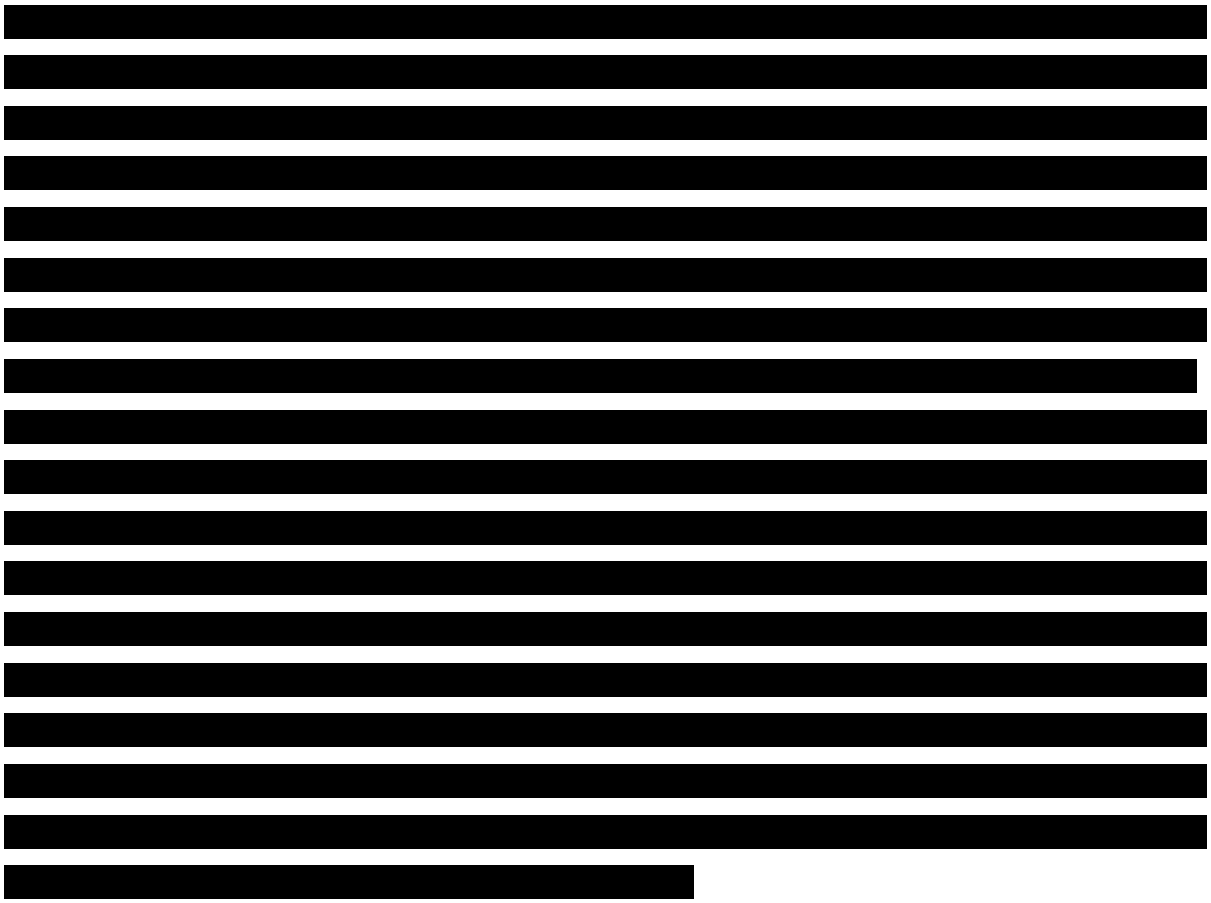
The YHEC review sets the scene and makes many useful points, including;

A more detailed table of faecal calprotectin tests included in the assessment can be found in the Diagnostics Assessment Report Addendum.

- The key issue is deciding which patients should be referred for endoscopic or radiological examinations. The usual definitive diagnosis is by colonoscopy and histology, but that it invasive, unpleasant, expensive, and with a risk, admittedly now small, of serious complications. There may also be long waiting lists. (p.7) One study reported that up to 40% of new GI referrals are for suspected IBS (p. 47)
- If a non-invasive test such as calprotectin could rule out IBD, patients would be spared endoscopy and might receive appropriate reassurance and treatment much earlier
- For this to happen, calprotectin needs to have a high negative predictive value, so that IBD can be ruled out.(p.8)
- Some patients with IBS do have biochemical evidence of inflammation (P5), and IBS may cover several subgroups. Some studies have reported higher calprotectin in patients with IBS, than in healthy controls, though the differences have not always been significant (P 20), and the levels in IBS are still well below a cut-off of 50 µg/g (p.25)
- The Rome criteria for diagnosing IBS may be met by many patients with organic disease, resulting in mis-diagnosis and failure to refer. (p.16)
- Faecal calprotectin is a marker of intestinal inflammation, not a test for organic versus non-organic disease (p.16)
- Most studies were from secondary care, and selection by GPs of patients likely to have organic disease may mean that results in secondary care may not be applicable to the different patient mix seen in primary care. (p.20)
- The YHEC report considered that high sensitivity was very important and that false positives were preferable to false negatives.
- The upper reference limit for absence of disease was suggested as 50 µg/g
- When using point of care tests, borderline or elevated results should be re-examined using a quantitative method.(p.22)
- Calprotectin was much better than blood tests, CRP and ESR, with NPVs 89%, 68% and 69% respectively. (p.27) The best blood test was CRP but it was effective in only 53% of patients (P48). In cost-effectiveness analysis, calprotectin dominated CRP and ESR (P55), giving more correct diagnoses at less cost.(Tables 32 and 33, p.55)

Further details of the YHEC and other reviews are given in

Table 5. Reporting of several aspects of the review was scanty. However it should be noted that the YHEC remit was restricted and did not include doing a full systematic review to standards such as Cochrane.



The review by van Rheenen et al appeared to match our main interest, since it was reported to be about the value of calprotectin in the investigation of suspected IBD, with a view to determining whether it reduced the number of unnecessary endoscopies.[47] It also appeared to be a high quality review. However not all the included studies were of newly presenting patients. Bunn 2001 had more patients with confirmed IBD than new patients.[63] Kolho 2012 enrolled a group of newly presenting patients but only 30 of the 132 stool samples were taken at presentation, with other being taken after treatment, as long as 72 weeks later.[64] So the patient group was correct, but timing of testing not suitable for our purposes.

One advantage of the van Rheenen review for our purposes was that overall, only 32% of adults with symptoms were found to have IBD. That proportion may be more similar to the mix of patients seen in primary care than some studies from specialist care. A disadvantage is that only two studies in adults excluded patients with rectal bleeding. Such patients would normally be referred for GI investigation on "red flag" grounds and so are outwith the remit of this review. However bleeding seems to be quite common in people with IBS. For example, Otten et al report that 26% of the group confirmed as having IBS, had rectal bleeding.[65] Other studies in the van Rheenen review mentioned rectal bleeding but did not give proportions.[66-68]

Van Rheenen et al reported that the pooled results gave sensitivity of 93% (95% CI 0.85 to 0.97) and specificity of 0.96 % (0.79 to 0.99). Screening by calprotectin would reduce the number of adults requiring endoscopy by 67%, but they estimated that 3% without IBD would have endoscopy and 2% with IBD would not be endoscoped and so have the disease missed.

However, they appeared to have pooled results at all cut-off levels, so that they pooled Schroeder 2007[69], which used a cut-off of 24 µg/g, with Limburg 2000[70], which used a cut-off of 100 µg/g. Such pooling does not seem appropriate.

The adult results reflect the high proportion with IBS. The results in children differ because only about 7% had IBS, and 61% had IBD. Van Rheenen et al estimated that the number requiring endoscopy would be reduced by only 35%, with 9% of those without IBD having endoscopy, and 5% with IBD being missed.

Van Rheenen et al noted that most studies were from secondary care, and provide a Fagan plot so that results for a population more representative of that seen in primary care can be estimated. From this, they expect that given a primary care expected prevalence of 5% with IBD, the NPV would be over 99.8%, good for ruling out IBD. However the PPV falls to 55%. (The cut-off level is not clear, since results are described as normal or not normal.)

Van Rheenen et al concluded that calprotectin is a useful test for identifying those most likely to need endoscopy.

An earlier review, by von Roon et al, had a broader remit, examining the value of calprotectin in the diagnosis of both IBD and colorectal cancer.[48] It was a high quality review. The approach was less suited to our purposes, since they included studies with healthy controls, and others in patients with known IBD. Some studies did not include people with IBS. Neverthless, some useful findings were that;

- The sensitivity of CRP was low, ranging from 35% to 40%
- The sensitivity of ESR was also low, 18% to 52%.
- For IBD, a cut-off of 100 µg/g gave slightly better precision than 50 µg/g, with areas under the curves of 0.98 and 0.95.
- Calprotectin at a cut-off of 50 µg/g performed well for differentiating between those with IBS and healthy controls, and those with IBD, AUC 0.97, with slightly higher precision at cut-off 100µg/g.

- Sensitivity for CD was higher than for UC (CD 0.95 in adults and 0.97 in children at 50 µg/g cut-offs, and UC 0.78)
- Levels of calprotectin in people with IBS were similar to those in healthy controls
- Calprotectin could not be recommended as a screening test for colorectal cancer.
- A sensitivity analysis excluding lower quality studies improved the sensitivity without affecting specificity, as did excluding smaller studies.

Von Roon et al also pooled results, but more correctly, pooling only studies using the same cut-offs. The pooling did not include the grouping we would have found most useful. They pooled IBD versus not IBD, and CD versus a mix of healthy controls and IBS. And most of the studies they included were not in newly presenting patients. The data below come from their Table 3.

Table 4. IBD versus non-IBD from Von Roon et al (2007)[48]

IBD versus non-IBD						
Group	**Cut-off (µg/g)**	**Patients**	**Studies**	**Se %**	**Sp %**	**AUC**
Adults and children	50	1267	9	89	81	0.95
Adults and children	100	328	4	98	91	0.98
Adults	50	1030	6	71	80	0.94
Children	50	201	3	83	85	0.96
Children	100	231	3	98	97	0.99
CD versus normal controls and IBS						
Adults	50	614	4	95	84	0.97
Children	50	119	2	97	79	-
Children	100	155	2	100	98	-
UC versus normal controls and IBS						
Adults and children	50	235	2	78	78	-

Note the suggestion that calprotectin may be less sensitive in UC than CD.

Von Roon et al noted some weaknesses in the evidence, including spectrum bias, commenting that; "*FC has a good diagnostic precision for separating IBD from non-IBD diagnoses overall. Whilst this finding is likely to hold true in patients with severe IBD, it may not necessarily translate to a clinical setting where the patient has a low pre-test probability of IBD, i.e. where a clinician is attempting to differentiate patients with functional abdominal pain syndromes or IBS from IBD patients with mild "functional-like" symptoms."*

Jellema et al set out to do a systematic review on the diagnosis of IBD in primary care, in adults only.[71] Their intention appears to have been to exclude studies in patients with established IBD. In order to increase relevance to primary care, they excluded studies in which the prevalence of IBD was

more than 25%, though as they point out, even that would be a high prevalence for a primary care population. (Though as noted above, we need to take into account the difference between the prevalence of IBD in the whole primary care population, and the prevalence in those selected by GPs for referral to specialist care.)

Unfortunately, few of their 24 included studies were carried out in primary care – only three partly in that setting. It was a high quality review. No pooling of results was done. Useful findings included;

- Symptoms associated with IBD (diarrhoea, abdominal pain, blood in stools, weight loss) provided individually poor sensitivity and specificity
- Amongst blood and faecal tests, calprotectin performed best
- The performance of CRP was very variable, with sensitivity ranging from 0.55 to 1.0 depending on cut-offs; specificity ranging from 0.42 to 0.90. ESR was similar.

Jellema et al had reservations about applying results from specialist care to primary care;
"*In a setting with low disease prevalence, the same combination of sensitivity and specificity will lead to much lower positive predictive values compared with a setting with a high disease prevalence."*

Kostakis et al reviewed the evidence on faecal calprotectin in paediatric IBD.[72] Few details of methods were given so the quality score was low. No data were given by type of control – which could be healthy children or "other GI disease". They included some studies with no controls. They concluded that the cut-off should be 50 µg/g rather than 100, on the basis of slightly higher sensitivity (95.8 to 100% versus 87 to 100%, after excluding an outlier study) but similar specificity (68 to 93 versus 69 to 94). No pooling of results was done.

The most recent systematic review comes from Henderson et al (2013) (personal communication – manuscript submitted for publication), and was of paediatric studies. It was a high quality review, enhanced by the contacting of authors for further information. This meant that they could include a study (Perminow 2009) which we did not [73]) after they obtained unpublished details. The selection was rigorous, with children required to have at least colonoscopy. This meant excluding a study (Van de Vijver 2012)[74]) where children with negative FCs did not have colonoscopy, but were instead followed up for 6 months. As will be reported below, we were less rigorous and allowed this to be included.

Henderson et al included 8 studies with a total of 715 subjects. Quality was assessed using a modified QUADAS checklist, with no studies achieving full marks, with spectrum bias being one problem, attributed to selection bias amongst referrals to tertiary centres. Most studies used a cut-off of 5o µg/g.

The authors concluded that FC testing had high sensitivity of almost 98%, with reasonable specificity of 68%. Positive LR was 3.07, negative LR 0.03. They noted that FC testing was inexpensive (their local cost being about £28 including labour costs).This compares with the cost of day case endoscopic assessment in children of £1500, and the additional costs of small bowel imaging.

Table 5. Characteristics and conclusions of previous reviews

Study	Inclusion criteria and methodology	Included studies	Conclusions
CEP 2010[62] **focus:** faecal calprotecting for distinguishing between inflammatory bowel disease and irritable bowel syndrome **Overall quality:** medium	**INCLUSION CRITERIA** **study design:** any **participants:** not explicitly defined, patients with possible inflammatory bowel disease or irritable bowel syndrome; **diagnostic procedure:** laboratory and point-of-care tests for faecal calprotectin and other inflammatory markers **outcomes:** sensitivity, specificity, positive predictive values, negative predictive values. **METHODOLOGY** **search strategy:** search of 11 databases (some not very relevant like CINAHL and BNI), studies published in the past 10 years; search terms indicated; English language only **study selection:** not reported **quality assessment:** not reported **data extraction:** not reported **data analysis:** text and tables	**number of included studies:** 43 (?) - search results not described **number of participants:** about 5050 **study quality:** not reported **participants:** not described in summary **diagnostic procedure:** cut-off values for faecal calprotectin ranged between 18.6 µg/g and 250 µg/g	**Conclusions:** Faecal calprotectin performs well in distinguishing organic bowel disease from functional bowel disease; sensitivity and specificity are over 80% in most studies (at cut-off 50 µg/g); where calculated, most positive and negative predictive values were 70 to 90% **Recommendations for practice:** none **Recommendations for research:** none

Study	Inclusion criteria and methodology	Included studies	Conclusions
Jellema 2011 **focus:** summary of diagnostic tests in patients with abdominal symptoms **Overall quality:** high	**INCLUSION CRITERIA** **study design:** cohort studies, case-control studies where controls were diagnosed with irritable bowel syndrome or in whom organic gastrointestinal disease was excluded **participants:** adult population consulting a physician because of non-acute gastrointestinal symptoms (primary care, open-access clinics, outpatient population with prevalence of IBD of 25% or less); target condition was IBD, but the perspective was from primary care, "Non-acute" was defined as symptoms for more than 2 weeks. **diagnostic procedure:** primary diagnostic studies; studies using colonoscopy, histology, barium enema and/or clinical follow-up to diagnose IBD (reference tests); index tests included: signs and symptoms, blood and faecal tests, abdominal ultrasonography [only faecal calprotectin considered here] **outcomes:** sensitivity, specificity, data for construction of two-by-two table. Studies were excluded in 2x 2 table could not be constructed. **METHODOLOGY** **search strategy:** MEDLINE, EMBASE for studies published up to Feb 2009; search terms indicated; search of reference lists of relevant articles and reviews etc.; languages restricted to English, Dutch, German, French **study selection:** selection by two independent authors; disagreements resolved by discussion; third author consulted in case of persisting disagreement **quality assessment:** yes, modified QUADAS tool **data extraction:** pre-tested forms; data extraction by two independent authors **data analysis:** diagnostic two-by-two tables, diagnostic performance measures; text and tables; distinguish between Crohn's disease and ulcerative colitis	**number of included studies:** 9 on faecal calprotectin **number of participants:** 863 **study quality:** 5 with positive assessment on 8 or more of 11 quality items; range 4 to 10; only a minority of studies used a design relevant to primary carr (and none of these were studies of calprotectin) **participants:** all primary diagnosis – appeared to be newly presenting patients **diagnostic procedure:** diagnostic cut-off points 15 µg/g and 170 µg/g, 10 to 30 mg/l	**Conclusions:** Calprotectin showed consistent and promising findings but none of the studies were performed in primary care. Authors conclusions: "Faecal calprotectin has excellent negative predictive value in patients with abdominal symptoms." **Recommendations for practice:** none **Recommendations for research:** Authors conclusions" "Before calprotectin can be used to guide clinical decisions in primary care, these markers need to be investigated by high quality prospective studies in that speicif setting".

Study	Inclusion criteria and methodology	Included studies	Conclusions
Kostakis 2012[72] **focus:** faecal calprotectin for diagnosis and confirming relapse in paediatric inflammatory bowel disease **Overall quality:** low	**INCLUSION CRITERIA** **study design:** primary studies; case reports excluded **participants:** patients aged ≤18 years with inflammatory bowel disease (IBD), both newly diagnosed and previously confirmed **diagnostic procedure:** measurement of faecal calprotectin **outcomes:** sensitivity, specificity, positive likelihood ratio (PLR), negative likelihood ratio (NLR) **METHODOLOGY** **search strategy:** MEDLINE, EMBASE for studies published up to Oct 2011; search terms indicated; English language only **study selection:** no details on study selection given **quality assessment:** no quality assessment **data extraction:** no details on data extraction given **data analysis:** text and tables; distinguish between IBD in general, Crohn's disease, ulcerative colitis, assessment at first diagnosis or to assess activity / relapse	**number of included studies:** 34 **number of participants:** 1345 with IBD (range 8 to 128), 1225 controls (range 0 to 509) **study quality:** not reported **participants:** 13 studies in newly diagnosed patients; 9 studies in patients under treatment; 10 studies including both . No data provided on type of controls, who could be healthy controls or "other GI disease", or have "functional disease" not specified. Two studies in newly diagnosed had no controls. **diagnostic procedure:** cut-off values for faecal calprotectin ranged between 50 µg/g and 275 µg/g	**Conclusions:** The faecal calprotectin test could be used for supporting diagnosis or confirming relapse of IBD in paediatric patients before they undergo GI endoscopy. A positive result could confirm the suspicion of either IBD diagnosis or IBD relapse (high sensitivity), but a negative result should not exclude these conditions (moderate specificity) **Recommendations for practice:** 50 µg/g of faecal calprotectin should be the cut-off point for detecting IBD **Recommendations for research:** none

Study	Inclusion criteria and methodology	Included studies	Conclusions
Van Rheenen 2010[47] **focus:** faecal calprotectin for investigation of suspected inflammatory bowel disease **Overall quality:**medium	**INCLUSION CRITERIA** **study design:** diagnostic accuracy studies, **participants: The authors state that** patients with inflammatory bowel disease suspected on clinical grounds, with previously diagnosed IBD were to be excluded. However at least one study, Bunn, was included despite most patients having previously confirmed IBD. Studies with healthy controls also excluded. **diagnostic procedure:** stool sampling (for faecal calprotectin, index test) before endoscopic evaluation including histopathological verification of segmental biopsies (reference standard) **outcomes:** sensitivity, specificity, positive likelihood ratio (PLR), negative likelihood ratio (NLR) **METHODOLOGY** **search strategy:** MEDLINE, EMBASE for studies published up to Oct 2009; search terms indicated; English language only; reference lists checked **study selection:** first selection by one reviewer; full text articles checked for eligibility by two independent reviewers; disagreements resolved by discussion. Selection based partly on having spectrum of patients relevant to question. **quality assessment:** QUADAS (7 most differentiating items), no details of duplicate assessment but looks to have been done thoroughly **data extraction:** items extracted were reported; no details of duplicate extraction **data analysis:** meta-analysis, ROC curves; text and tables; distinguish between adults and children	**number of included trials:** 13 **number of participants:** 670 adults, 371 children / adolescents **trial quality:** studies in children / adolescents were better quality than studies in adults; 1 study fulfilled all 7 criteria, 4 fulfilled 6 of 7, 4 fulfilled 5 of 7, 2 fulfilled 4 of 7, and one each 3 and 2 of 7. All studies reported FC followed by endoscopy. **participants:** 6 studies in adults, 7 in children / adolescents; prevalence of IBD between 14 and 80% (32% of adults, 61% of children / adolescents); all studies were from hospital clinicsl **diagnostic procedure:** cut-off values for faecal calprotectin ranged between 24 µg/g and 150 µg/g	**Conclusions:** Testing for faecal calprotectin is a useful tool for identifying patients who are most likely to need endoscopy for suspected inflammatory bowel disease; the discriminatory power to safely exclude inflammatory bowel disease was significantly better in studies of adults than in studies of children; at a tertiary care level, faecal calprotectin can contribute important information In adults, an abnormal FC result gave 91% probability of IBD and a normal one a 3% probability. **Recommendations for practice:** the authors reserved judgment about the utility of faecal calprotectin in primary care , given the lack of studies in primary care. **Recommendations for research:** none

Study	Inclusion criteria and methodology	Included studies	Conclusions
Von Roon 2007[48] **focus:** diagnostic precision of faecal calprotectin for inflammatory bowel disease and colorectal cancer in adults and children **Overall quality:** high	**INCLUSION CRITERIA** **study design:** diagnostic studies with a control group **participants:** patients with Crohn's disease, ulcerative colitis or colorectal cancer compared with healthy patients or those with irritable bowel syndrome **diagnostic procedure:** faecal calprotectin compared with histological diagnosis **outcomes:** sensitivity, specificity, area under the summary ROC curve, diagnostic odds ratio **METHODOLOGY** **search strategy:** MEDLINE, EMBASE, Cochrane Library for studies published up to March 2006; search terms indicated; no language restrictions; reference lists checked; funnel plot suggested no publication bias. **study selection:** not reported **quality assessment:** QUADAS, no details of duplicate assessment **data extraction:** data extracted independently by two authors, in case of disagreement, consensus was reached through discussion with the senior author **data analysis:** meta-analysis, ROC curves; heterogeneity assessment; text and tables; distinguish between adults and children, IBD in general, Crohn's disease, ulcerative colitis, colorectal neoplasia	**number of included trials:** 30 **number of participants:** 5983 **trial quality:** 19 studies rated high quality (QUADAS score above 11); range 10 to 13 **participants:** 22 studies in adults, 1 in adults and children, 7 in children **diagnostic procedure:** 2 studies assessed diagnostic precision in predicting relapse and 3 in examining disease activity; cut-off values for faecal calprotectin ranged between 18.6 µg/g and 250 µg/g Results Sensitivity analyses showed that high quality studies (QUADAS >11) had higher Se – 0.90 versus 0,71 (adults, 50ug, IBD vs no IBD) when all studies included; no different in Sp. Large studies (>100) also gave higher Se,	**Conclusions:** faecal calprotectin cannot be recommended as a screening test for colorectal cancer in the general population; faecal calprotectin appeared to offer a good diagnostic precision in distinguishing inflammatory bowel disease from non-IBD diagnoses with a higher precision at a cut-off of 100 µg/g. FC in patients with IBS was no different from in healthy controls. FG was better for CD than UC, and better in children **Recommendations for practice:** none **Recommendations for research:** high quality study needed investigating different cut-off points for faecal calprotectin

Study	Inclusion criteria and methodology	Included studies	Conclusions
Henderson 2013 (pers. comm.) **Focus:** the value of FC testing in chldren being investigated for suspected IBD **Overall quality:** high	**INCLUSION CRITERIA** **study design:** retrospective or prospective case-control studies **participants:** children with suspected bowel inflammation (PIBD) who underwent at least colonoscopy **diagnostic procedure:** faecal calprotectin compared against ileocolonoscopy or upper endoscopy **outcomes:** sensitivity, specificity, PLR, NLR, ROC curve **METHODOLOGY** **search strategy:** MEDLINE was searched up to May week 3 2012 EMBASE up to week 25 2012. PubMed, Google Scholar and the Cochrane Library were searched; search strategy available on request; reference list checked, personal collections and meeting abstracts were checked (only full text articles were included); no language restrictions (foreign language articles were translated using Google Translate) **study selection:** studies evaluated by two reviewers independently for eligibility, any discrepancies were resolved by discussion, **quality assessment:** modified version of the QUADAS tool (11 questions) **data extraction:** data entered into a customised database. Authors were contacted if certain parameters were uncertain mainly during the construction of 2x 2 table; **data analysis:** meta-analysis, 2x2 table, sensitivity, specificity, ROC curve, no analysis of heterogeneity.	**number of included trials:** Eight **number of participants:** 715 (394 PIBD patients and 321 non-PIBD) **trial quality:** one study each fulfilled 9, 8 and 6 criteria respectively; two studies fulfilled 5 criteria while three studies fulfilled 2 criteria; in the studies that did not fulfil most criteria had most items unclear. Only three studies had representative spectrum of patients. **participants:** paediatric patients with suspected IBD. More had CD than UC: ratio CD:UC about 1.5:1 **diagnostic procedure:** in six studies, the cut-off value was 50µg/g whereas in two studies, the cut-off was 100µg/g	**Conclusions:** faecal calprotectin is a useful tool to screen children with suspected bowel inflammation. The test may lower endoscopy rates thereby benefiting both parents and children. **Recommendations for practice:** **Recommendations for research:** studies to see if FC testing reduces endoscopy rates and assess cost benefits, and studies of the usefulness of FC in disease monitoring.

Abbreviations: IBD – inflammatory bowel disease, NLR – negative likelihood ratio, PLR – positive likelihood ratio; IBDU – inflammatory bowel disease type unclassified; PIBD - paediatric inflammatory bowel disease

Table 6. Quality of previous reviews

Review	Clear definition of review question (PICOS)	Search strategy adequate and appropriate	Minimisation or error and bias in study selection	Appropriate quality assessment of included studies (e.g. QUADAS)	Appropriate data extraction process	Sufficient detail on primary studies	Appropriate methods used for data synthesis and comparison between studies	Conclusions reflect evidence reviewed
CEP 2010	no	yes	not reported	not reported	not reported	yes	unclear	yes
Jellema 2011	yes	yes	yes	yes	yes	yes	yes	yes
Kostakis 2012	yes	partially	not reported	no	not reported	partially	yes	yes
Van Rheenen 2010	yes	partially	partially	yes	yes	yes	yes	yes
Von Roon 2007	yes	yes	not reported	yes	yes	yes	yes	yes
Henderson 2013	yes	yes	yes	yes	yes	yes	yes	yes

1 to 4 criteria met: low quality; 5 to 6 criteria met: medium quality; 7 to 8 criteria met: high quality

Table 7. Results of previous reviews

Review	**Results** (sensitivity is for diagnosing IBD)
CEP 2010	**Sensitivity:** 63 to 100% **Specificity:** 37 to 100% **Positive predictive value:** 60 to 100% **Negative predictive value:** 51 to 100%
Jellema 2011	**Sensitivity:** 84 to 100% in 7 studies, 61 and 64% in 2 studies **Specificity:** 71 to 100%
Kostakis 2012	**Newly diagnosed and untreated IBD:** **ALL** **Sensitivity:** 73.5 to 100% (95.8 to 100% for 50 µg/g as cut-off point, 73.5 to 100% for 100 µg/g as cut-off point) **Specificity:** 65.9 to 100% (65.9 to 92.9% for 50 µg/g as cut-off point, 69.2 to 100% for 100 µg/g as cut-off point) **PLR:** 2.8 to 34.9 (2.9 to 14 for 50 µg/g as cut-off point, 2.8 to 34.9 for 100 µg/g as cut-off point) **NLR:** 0 to 0.3 (0 for 50 µg/g as cut-off point, 0 to 0.3 for 100 µg/g as cut-off point) **ULCERATIVE COLITIS**

Review	**Results** (sensitivity is for diagnosing IBD)
	Sensitivity: 75 to 100% **Specificity:** 65.9 to 92.9% **PLR:** 2.4 to 14 **NLR:** 0 to 0.4 **CROHN'S DISEASE** **Sensitivity:** 93.3 to 100% **Specificity:** 65.9 to 92.9% **PLR:** 2.9 to 14 **NLR:** 0 to 0.1 **Already diagnosed and under treatment IBD:** **Sensitivity:** 12.5 to 100% (100% for 50 µg/g as cut-off point, 12.5 to 68.2% for 100 µg/g as cut-off point) **Specificity:** 58.3 to 100% (58.3 to 80% for 50 µg/g as cut-off point, 69.2 to 100% for 100 µg/g as cut-off point) **PLR:** 1.1 to 5 (2.4 to 5 for 50 µg/g as cut-off point, 1.1 for 100 µg/g as cut-off point) **NLR:** 0 to 1 (0 for 50 µg/g as cut-off point, 0.9 to 1 for 100 µg/g as cut-off point) Faecal calprotectin levels are much higher in patients with active IBD (newly diagnosed without treatment of under treatment with relapse) than in patients with IBD in remission, but faecal calprotectin levels in patients with inactive IBD are higher than those of healthy controls or patients with functional disorders or other GI diseases
Von Rheenen 2010	**Adults:** **Sensitivity:** 93% (95% CI 85 to 97) **Specificity:** 96% (95% CI 79 to 99) **PLR:** 20 **NLR:** 0.06 **Children / adolescents:** **Sensitivity:** 92% (95% CI 84 to 96) **Specificity: 7**6% (62% CI 79 to 86) **PLR:** 5 **NLR:** 0.1
Von Roon 2007	**Adults and children (cut-off 50 µg/g):** **Sensitivity:** 89% (95% CI 86 to 91) **Specificity:** 81% (95% CI 78 to 84)

Review	**Results** (sensitivity is for diagnosing IBD)
	<u>Adults and children (cut-off 100 µg/g):</u> **Sensitivity:** 98% (95% CI 93 to 99) **Specificity:** 91% (95% CI 86 to 95) The diagnostic precision was higher in children than adults and at a cut-off of 100 versus 50 µg/g In adults, cut-off 50ug, UC Se 0,78m Sp 0,78 CD adults, Se 0,95 and Sp 0,85 at 50 Children with CD, 0.97 and 0.79 at 50; 1.0 and 09,8 at 100
Henderson 2013	**<u>Pooled sensitivity and specificity</u>** **Sensitivity:** 0.978 (95% CI 0.947-0.996) **Specificity:** 0.682 (95% CI 0.502-0.863) **Positive likelihood ratio:** 3.07 **Negative likelihood ratio:**0.03

Summary

Some reviews are now out of date. The most recent ones (YHEC, Jellema, Henderson) all conclude that faecal calprotectin testing is very useful. Henderson focuses only on use in children but is right up to date, and very high quality.

Table 8 has been updated. Please refer to the Diagnostics Assessment Report Addendum

2.3 The tests

Table 8 shows the calprotectin tests included, and the studies of each included in sections 2.5 to 2.8. Note that the numbers of studies apply only to those that we could include in our meta-analyses. There are other studies of these tests, and indeed we include some elsewhere in this report.

Table 8. Evidence base for the calprotectin tests.

Name of Test	Type of test	Evidence base
Nycomed	ELISA	IBS vs. IBD: One study, El Badry 2010 IBD vs. non-IBD: 2 studies; Limberg 2000; Sidler 2008 Organic vs. IBS: none Organic vs. non-organic: none
Immundiagnostic ELISA kit	ELISA	IBS vs. IBD: 2 studies, Basumani 2013; Schroder 2007. IBD vs. non-IBD: none Organic vs. IBS: 1 study, Basumani 2013 Organic vs. non-organic: none
EK-CAL	ELISA	IBS vs. IBD: none IBD vs. non-IBD: 1 study, Damms 2008 Organic vs. IBS: none Organic vs. non-organic: 3 studies
Calprest	ELISA	IBS vs. IBD: none IBD vs. non-IBD: 5 studies: Fagerberg 2005; Diamanti 2010; Tomas 2007; Canani 2006; licata 2012 Organic vs. IBS: 1 study Organic vs. non-organic: 3 studies
CALPRO Calprotectin ELISA test (ALP)	ELISA	IBS vs. IBD: Otten 2009; Schoepfer 2008; Li 2006 IBD vs. non-IBD: Vijfer 2012; Henderson 2012 Organic vs. IBS: none Organic vs. non-organic: none
Not known	ELISA	IBS vs IBD: Bharathi 2005 IBD vs non-IBD: Ashorn 2009
Quantum Blue	POCT	IBS vs. IBD: none IBD vs. non-IBD: none Organic vs. IBS: none Organic vs. non-organic: 1 study
Prevent ID Caldetect	POCT	IBS vs. IBD: 1 study IBD vs. non-IBD: none Organic vs. IBS: none Organic vs. non-organic: 1 study
Prevista(no longer available)	POCT	IBS vs. IBD: none IBD vs. non-IBD: 1 study Organic vs. IBS: none Organic vs. non-organic: none
EliA platform	EliA	None

2.4 The comparisons

The decision problem concerns the use of calprotectin to help distinguish between inflammatory and non-inflammatory bowel conditions. For GPs, this is part of distinguishing between patients who need to be referred to secondary care, and those who can be managed in primary care. However in practice, the distinction in adults is usually between patients at the more troublesome end of the IBS spectrum and IBD, we start with that in section 2.5. In adult medicine, this is the most important comparison. In section 2.6 we look at another way of distinguishing between patients who should be referred, and those with IBS, in two studies that compare "organic" with IBS. In adult medicine there are other organic causes that can cause symptoms such as colorectal neoplasia.

Note that we are assuming that in the situations in which calprotectin would be used, coeliac disease has already been detected or ruled out by blood testing. It is a bowel disease characterised by inflammation, but would not have high calprotectin levels because the inflammation is mediated by lymphocytes, not neutrophils. In children with coeliac disease, calprotectin may be mildly elevated.

In paediatrics, studies aim to distinguish between IBD and non-IBD, since IBS is much less common. Some adult studies also make this comparison, but most studies of IBD versus non-IBD come from paediatric gastroenterology. These are dealt with in section 2.7.

In section 2.8, we include "organic versus non-organic". This comparison is less relevant, because the organic group can contain a mixture of inflammatory and non-inflammatory conditions. We deal with this group in less detail.

2.5 Studies of calprotectin in the differentiation of IBD and IBS

We included seven studies in this group, shown in the paired forest plots below (Figure 3). One of these studies (Basumani 2012) is not yet published and is [redacted]. One study (Bharathi 2005), available only as an abstract, gave no detail of clinical setting. Only 3 studies gave data at cut-offs other than 50 µg/g, and one did not provide enough data to calculate sensitivity. As expected, low thresholds gave high sensitivity for IBD but poor specificity. The studies were in adults.

All used ELISA tests, and one[65] also used a POCT test.
Note that numbers in the tables reflect total numbers in each study, and not all may be relevant for our purposes. Numbers in forest plots will sometimes be smaller than numbers in the studies. For example some studies included "healthy controls" who are not relevant to this review.

Table 9. Outline of studies comparing IBD versus IBS

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Schroder 2007[69]	76	Diarrhoea for more than 4 weeks, unknown cause unknown	hospital	To assess utility of FC to detect inflammation in patients with ? IBD, ? IBS	Colonoscopy and biopsy	Previous investigations, GI bleeding, polyps, pregnancy
Otten 2008[65]	114	Consecutive patients referred with lower abdominal symptoms, referred to endoscopy unit	Endoscopy unit, Netherlands	To evaluate POCT FC and lactoferrin tests for assessing inflammation; and differentiating IBS and IBD	Colonoscopy and biopsy	Age under 18; previous colon surgery; iron deficiency
Schoepfer 2008[67]	94	Outpatients and inpatient	Gastroenterology Department, University Hospital Switzerland	To assess accuracy FC and lactoferrin to detect inflammation in patients with ?IBS, ?IBD	Colonoscopy including terminal ileum and biopsies	Incomplete colonoscopy, microscopic colitis, no FC sample, infections, polyps, aspirin or NSAIDs, etc
El-Badry 2010[76]	29	GI symptoms for at least 6 months, and endoscopy necessary to exclude organic pathology	Internal Medicine Department, Cairo	To evaluate FC at different cut-offs for differentiation functional and organic disorders	Colonoscopy into ileum with biopsies	NSAIDS, aspirin, anticoagulants, arthritis and other diseases affecting FC
Li 2006[55]	240	Outpatients and inpatients with IBS or IBD, healthy controls; patients followed up after polyp removal with no recurrence	Hospital, Peking	To assess FC in differential diagnosis of IBS and exclude organic diseases	Colonoscopy with biopsy in IBD group	No upper GI symptoms; adenomas; severe other disease
Bharathi 2005[77]	58	Patients presenting with abdo pain or loose stools	Not reported	To assess NPV of FC for excluding bowel pathology in young patients with ? IBS	Various – endoscopy, ultrasound	Not reported

Table 10. QUADAS quality assessment of studies comparing IBD versus IBS

	Basumani 2012	Schroder 2007	Otten 2008	Schoepfer 2008	El-Badry 2010	Li 2006	Bharathi 2005

Spectrum	[redacted]	Yes	Yes	Yes	Yes	Unclear	Yes
Reference standard	[redacted]	Yes	Yes	Yes	Yes	Yes	Yes
Acceptable delay?	[redacted]	Yes	Yes	Yes	Yes	Yes	Unclear
Whole sample verified?	[redacted]	Yes	No	Yes	Yes	Yes	No
Same reference standard	[redacted]	Yes	Unclear	Yes	Yes	Unclear	Yes
Newly diagnosed?	[redacted]	Yes	Yes	Yes	Yes	Unclear	Yes
Blinded reference testing?	[redacted]	Yes	Yes	Yes	Unclear	Unclear	Unclear
Index results blinded?	[redacted]	Yes	Yes	Yes	Unclear	Unclear	Unclear
Same clinical data	[redacted]	Yes	Yes	Yes	Unclear	Yes	Unclear
Intermediate results reported?	[redacted]	Yes	Yes	Yes	Yes	Yes	Yes
Withdrawals explained?	[redacted]	Yes	yes	yes	Yes	unclear	yes

Table 11. Results of studies comparing IBD versus IBS

Study	**Cut-off value** µg/g	**PPV (95% CI)**	**NPV (95% CI)**	**PLR (95% CI)**	**NLR (95% CI)**	**Accuracy**	**Disease Prevalence % (95% CI)**
[redacted]							

Study	Cut-off value µg/g	PPV (95% CI)	NPV (95% CI)	PLR (95% CI)	NLR (95% CI)	Accuracy	Disease Prevalence % (95% CI)
[redacted]		[redacted]	[redacted]	[redacted]	[redacted]		[redacted]
Bharathi 2005	60	0.00 (0.00 to 0.27)	1.00 (0.92 to 1.00)			0.79	0.00 (0.00 to 6.16)
El-Badry 2010	50	1.00 (0.81 to 1.00)	0.75 (0.43 to 0.95)		0.15 (0.05 to 0.43)	0.90	68.97 (49.17 to 84.72)
El-Badry 2010	100						
Li 2006	50	0.95 (0.86 to 0.99)	0.93 (0.84 to 0.98)	18.67 (6.18 to 56.63)	0.07 (0.03 to 0.18)	0.94	50.00 (40.74 to 59.26)
Otten 2008	≥15	0.82 (0.63 to 0.94)	1.00 (0.96 to 1.00)	18.2 (7.7 to 42.7)	0	0.96	20.18 (13.24 to 28.72)
Otten 2008	≥60	0.88 (0.60 to 0.98)	0.91 (0.83 to 0.96)	27.7 (6.7 to 113.3)	0.4 (0.2 to 0.7)	0.90	20.18 (13.24 to 28.72)
Otten 2008	>50	0.65 (0.47 to 0.81)	0.99 (0.93 to 1.00)	7.25 (4.25 to 12.38)	0.05 (0.01 to 0.34)	0.89	20.18 (13.24 to 28.72)
Schoepfer 2008	50	1.00 (0.93 to 1.00)	0.73 (0.57 to 0.86)		0.17 (0.10 to 0.29)	0.88	68.09 (57.67 to 77.33)
Schroder 2007	15	1.00 (0.92 to 1.00)	0.91 (0.76 to 0.98)		0.07 (0.02 to 0.20)	0.96	59.21 (47.33 to 70.35)

The most useful study was that of Basumani et al (see Figure 4), because it provided data at six cut-offs, as shown for clarity below. At the lower levels, as expected, sensitivity is high, but specific low.

Figure 3. IBD vs IBS

Figure 4. IBD vs IBS - Basumani data only

Figure 5. IBD vs IBS – Basumani data only

Figure 6. Summary receiver-operating characteristic (sROC) curve for fecal calprotectin in the diagnosis of bowel diseases IBD versus IBS at a cut-off level of 50 µg/g

The summary point shows the summary sensitivity, and the confidence contour shows the confidence interval or region for the summary point.

The prediction contour outlines the prediction region for the true sensitivity and specificity in a future study. (For details see Harbord and Whiting[49])

Figure 7. Forest plots of pooled sensitivity and specificity of fecal calprotectin in the diagnosis of bowel diseases IBD versus IBS at a cut-off level of 50 µg/g

It should be noted that some experts advise caution in the use of the I squared test for indicating heterogeneity in reviews of diagnostic test accuracy and suggest that they should not be routinely used (see Cochrane Handbook for systematic reviews of diagnostic accuracy V1.0 December 2012, pare 10.4.3)[50]

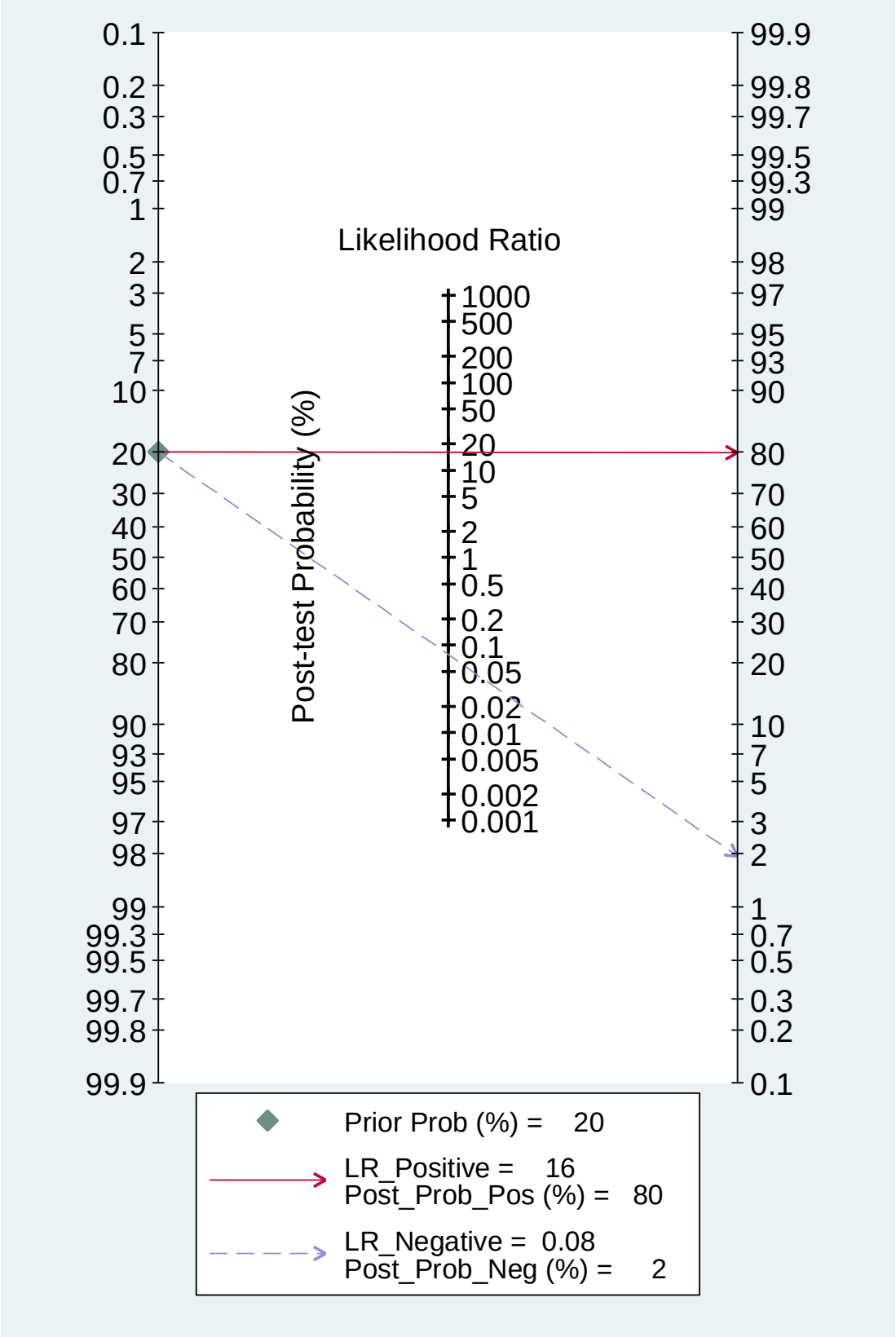


Figure 8. The use of the Fagan's nomogram (a straight line through the pre-test probability of 20% and the LR− of 0.20 yields a post-test probability of about 2%). [IBD versus IBS at a cut-off level of 50 µg/g].

Table 12. Diagnostic Odds Ratios - IBD vs IBS

Number of Patients	Number of Studies	Cut-off level (µg/g)	DOR (95% CI)
[redacted]	[redacted]	[redacted]	[redacted]
76	1	15	10 (4 to 27)
[redacted]	[redacted]	[redacted]	[redacted]
596	5	50	202 (47 to 868)
[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]

Note the large confidence intervals around all the DORs.

Conclusions of section 2.5

Calprotectin testing appears very useful for differentiating between IBS and IBD.

Almost all sensitivities are high, the outlier being Otten 2008 with the 60 µg/g cut-off using a POCT. As expected, and shown best by the Basumani data in Figure 4, there is a trade-off between sensitivity and specificity.

The only POCT test in the group is the Prevent ID, which performed well at the 15 µg/g cut-off, but not so well at the 60 µg/g one. Though it is curious that its specificity should be so high at the lower cut-off. The POCT with a 15 µg/g cut-off had higher specificity (95%) than the ELISA at 50 µg/g (87%).

The variability amongst sensitivities was much less than amongst specificities. Heterogeneity was moderate for Se (I^2 37%) but high for Sp (94%). (However see earlier note about the I^2 test in diagnostic reviews.) Even using the same PhiCal ELISA test with the 50µg/g cut-off, sensitivities ranged from 83% to 96%.

Figure 6 provides the summary: Se 93% and Sp 94%, for ELISA tests, at 50 µg/g cut-off. These are based on 5 studies. There was only one study for the 100 µg/g cut-off. With an AUC of 0.97 at 50µg/g, there is little room for improvement.

The only study using a POCT performed well at cut-off 15 µg/g with sensitivity 100% and specificity 95%. At 60 µg/g, sensitivity was only 61%, which is unlikely to be acceptable given the importance of not missing people with IBD.

All studies were on adults.

On this evidence base, it may be unwise to recommend any ELISA cut-off other than 50 µg/g.

2.6 Studies of calprotectin: organic versus IBS

The term organic covers a range of conditions, and the range varies amongst studies. Some of these conditions would not normally be regarded as inflammatory. Inflammation implies the presence of white blood cells, and one would not expect these in lesions such as colonic polyps. However, FC is often raised in patients with larger polyps, as shown in Figure 20, later.

So FC may flag up the presence of conditions other than IBD, such as some colorectal cancers and large adenomas, but results are more variable than with IBD. Therefore in studies with a mix of organic conditions, calprotectin may not appear as reliable. However, this should not detract from its good performance in detecting IBD and excluding IBS.

The low sensitivity in the Carroccio study may be partly due to their case mix, which is related to their institution's role as a referral centre for food intolerances. Their organic group included many (about a third) with coeliac disease who had negative calprotectins (because the inflammation is mainly lymphocytic).

Table 13. Outline of studies comparing organic versus IBS

Study	Number of patients	Recruits	Setting	Aim
Carroccio 2003	120	Chronic diarrhoea for more than 4 weeks, with or without abdominal pain; unknown origin	Outpatient clinics of the University Hospital and of the Pediatric Division of "Di Cristina" Hospital, Italy	To assess value of FC in ident

Table 14. QUADAS quality assessment of studies comparing organic versus IBS

	Carroccio 2003
Spectrum	Yes
Reference standard	Yes
Acceptable delay?	Yes
Whole sample verified?	No
Same reference standard	Yes
Newly diagnosed?	Yes
Blinded reference testing?	Yes
Index results blinded?	Yes
Same clinical data	Yes
Intermediate results reported?	Yes
Withdrawals explained?	Yes

Table 15. Results of studies comparing organic versus IBS

Study	Cut-off value	PPV (95% CI)	NPV (95% CI)	PLR (95% CI)	NLR (95% CI)	Accuracy	Disease Prevalence % (95% CI)
[redacted]							
[redacted]							
[redacted]							
[redacted]							
[redacted]							
[redacted]							
Carroccio 2003 (all patients)[78]	50 µg/g	0.83 (0.70 to 0.92)	0.83 (0.70 to 0.92)	4.04 (2.17 to 7.53)	4.04 (2.17 to 7.53)	0.74	54.17 (44.83 to 63.29)
Carroccio 2003 (adults)[78]	50 µg/g	0.70 (0.50 to 0.86)	0.74 (0.59 to 0.86)	3.17 (1.61 to 6.23)	0.46 (0.28 to 0.75)	0.73	42.86 (31.09 to 55.25)
Carroccio 2003 (children)[78]	50 µg/g	0.96 (0.80 to1.00)	0.58 (0.37 to 0.78)	10.71 (1.59 to 72.00)	0.31 (0.18 to 0.53)	0.78	70.00 (55.39 to 82.14)
Carroccio 2003 (all patients) [78]	100 µg/g	0.88 (0.73 to 0.97)	0.59 (0.48 to 0.70)	6.35 (2.38 to 16.90)	0.58 (0.46 to 0.74)	0.68	54.17 (44.83 to 63.29)
Carroccio 2003 (adults)[78]	100 µg/g	0.81 (0.54 to 0.96)	0.69 (0.55 to 0.81)	5.78 (1.81 to 18.48)	0.61 (0.44 to 0.85)	0.71	42.86 (31.09 to 55.25)
Carroccio 2003 (children)[78]	100 µg/g	0.95 (0.74 to 1.00)	0.45 (0.27 to 0.64)	7.71 (1.13 to 52.66)	0.52 (0.36 to 0.75	0.64	70.00 (55.39 to 82.14)

Figure 9. Organic vs IBS

Figure 10. Organic vs IBS - Basumani data only

Figure 11. Organic vs IBS – Basumani data only

Table 16. Diagnostic Odds Ratios - organic vs IBS

Number of Patients	Number of Studies	Cut-off level (µg/g)	DOR (95% CI)
[redacted]	[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]	[redacted]
239	2	50	3.3 (2.2 to 4.7)
[redacted]	[redacted]	[redacted]	[redacted]
239	2	100	2.7 (2.0 to 3.6)
[redacted]	[redacted]	[redacted]	[redacted]

2.7 Studies of calprotectin: IBD versus non-IBD.

There were 11 studies in this group, 8 in paediatrics and 3 in adults. All used ELISA tests, and one (Damms 2008) also used the Prevista POCT.

Details of the studies are in Table 17. It should be borne in mind that symptoms of IBD in children may be "subtle and atypical" (Sidler 2008) rather than the typical diarrhoea, abdominal pain and weight loss. Impaired growth can be one presentation.

Table 17. Outline of studies comparing IBD versus non-IBD

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
Ashorn 2009[79]	55	Suspicion of IBD	Hospital, Finland	To examine the association of FC with serological markers in children and adolescents with IBD	Upper and lower endoscopies with biopsy	Not reported
Canani 2006[60]	45	Suspicion of IBD	Paediatric Gastroenterology Unit, Italy	To assess the diagnostic accuracy of FC and other tests independently and in combination; to assess value of FC as screening tool for IBD	Presence or absence of previously reported clinical, radiographic, endoscopic and histopathologic criteria. Bowel wall ultrasonography within 24 hours	Patients with symptoms or signs strongly suggestive of IBD, such as abdominal mass..

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
				in children	of admission.	
Damms 2008[53]	84	Patients aged >18 years undergoing colonoscopy for GI disorders or for CRC screening – medical check-up	Gastroenterological departments of 3 hospitals and 3 outpatient Gastroenterology clinics, Germany	To assess the diagnostic accuracy of the new calprotectin rapid test compared to ELISA in detecting colonoscopy proven IBD and malignancies	Colonoscopy; for CRC screening – medical check-up.	Known extraintestinal inflammatory disease; NSAIDs; anticoagulants
Diamanti 2010[80]	197	Recurrent abdominal pain and altered bowel habit	Gastroenterology and Nutrition Unit of Hospital, Rome	To assess the diagnostic precision and value as a screening tool of FC compared to histology	Colonoscopy including intubation of terminal ileum with biopsy	Not reported
Fagerberg 2005[81]	36	Children with gastrointestinal symptoms who were scheduled for colonoscopy to rule out IBD	Hospitals in Stockholm, and Vasteras (Sweden)	To determine if FC can be used as a diagnostic test of colonic inflammation to identify those children who require colonoscopy.	Complete ileocolonoscopy with biopsy	Had no bacterial gastroenteritis detectable by faecal culture or serology and did not have any other chronic inflammatory disease.
Henderson 2012[26]	190	Patients undergoing endoscopy	The paediatric gastroenterology department ,Royal Hospital for Sick Children in Edinburgh	To describe the differences in FC levels between IBD types and non-IBD disease categories.	IBD patients: standard clinical, histological and radiological findings Non-IBD (control) patients: upper and lower endoscopy	Insufficient stool sample; aged <1 or >18 years; >6 months delay between sample and endoscopy; previous endoscopy; FC sample after endoscopy; known diagnoses of GI diseases
Licata 2012[56]	346	Consecutive patients referred with chronic (≥4 week) non-bloody diarrhoea of unknown	Gastroenterology outpatient department, University of Palermo	To assess the diagnostic performance of FC as a stool-screening biomarker for organic	Colonoscopy with biopsies	GI bleeding, known malignancies of bowel; colonic surgery; recent respiratory or UT infection;

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
		origin		intestinal disease		pregnancy, alcohol abuse, NSAIDs
Limburg 2000[70]	110	Patients referred for colonoscopy with chronic diarrhoea (≥4 weeks) of unknown origin or chronic colitis of unknown cause	The Mayo Clinic (Rochester, MN)	To assess and compare calprotectin and haemoglobin as stool screening biomarkers for colorectal inflammation	Colonoscopy; biopsies taken when clinically indicated	Abnormalities on GI x-rays; GI bleeding; GI endoscopy within the preceding 2 wk, epistaxis within the preceding 1 wk, active menstruation, known colorectal neoplasia, familial adenomatous polyposis, and hereditary nonpolyposis colorectal cancer syndrome.
Sidler 2008[82]	61	Children aged between 2 and 18 years referred for further investigation with GI symptoms suggestive of an organic bowel disease	Gastroenterology Outpatient Clinic at Sydney Children's Hospital, Australia	To define the appropriate roles for faecal S100A12 and calprotectin in the initial investigations of children with suspected IBD	Upper GI endoscopy and complete ileocolonoscopy with biopsy	Previous diagnosis of organic bowel disease; infectious gastroenteritis; use of NSAIDs, antibiotics, or corticosteroids preceding 2 weeks
Tomas 2007[61]	43	Patients referred for further investigation with GI symptoms	Paediatric Gastroenterology Unit of University Hospital, Spain	To evaluate FC in paediatric patients with signs and symptoms suggestive of IBD	Clinical criteria, laboratory, image and endoscopic test results	Not reported
van de Vijver 2012[74]	117	Children aged between 6 and 18 years referred for further investigation with abdominal symptoms and clinical suspicion of IBD	Paediatric outpatient clinics of six general hospitals and one tertiary care hospital in the northern region of the Netherlands.	To determine a diagnostic strategy to minimise the number of patients with negative endoscopy results without missing any cases of IBD	Some patients endoscopy; others – stool tests for bacteria, ova and parasites, gastroscopy, different imaging and dietary assessment. Complete	Younger children (who have higher normal values of FC)

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
					resolution after 6 months follow-up.	

Table 18. QUADAS quality assessment of studies comparing IBD versus non-IBD

	Ashorn 2009	Canani 2006	Damms 2008	Diamanti 2010	Fagerberg 2005	Henderson 2012	Licata 2012	Limburg 2000	Sidler 2008	Tomas 2007	van de Vijver 2012
Spectrum	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Yes	Unclear	Yes
Reference standard	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Acceptable delay?	Unclear	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Yes	Unclear	Yes
Whole sample verified?	Yes	Yes	Yes	Yes	Unclear	Yes	Yes	No	Yes	Yes	No
Same reference standard	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Yes
Newly diagnosed?	Yes	Yes	Unclear	Yes	Yes	Yes	Unclear	Yes	Yes	Yes	Yes
Blinded reference testing?	Unclear	Yes	Unclear	Yes	Yes	Unclear	Yes	Yes	Unclear	Unclear	Yes
Index results blinded?	Unclear	Yes	Yes	Yes	Yes	Unclear	Yes	Yes	Yes	Unclear	Yes
Same clinical data	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Intermediate results reported?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Yes
Withdrawals explained?	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes

Table 19. Results for studies comparing IBD versus non-IBD

Study	Cut-off value (µg/g)	PPV (95% CI)	NPV (95% CI)	PLR (95% CI)	NLR (95% CI)	Accuracy	Disease Prevalence % (95% CI)
Ashorn 2009	≥100	0.96 (0.89 to 1.00)	0.67 (0.38 to 0.88)	9.75 (1.50 to 63.37)	0.13 (0.05 to 0.29)	0.89	80.00 (67.03 to 89.57)
Canani 2006	95.3	0.93 (0.76 to 0.99)	0.89 (0.65 to 0.99)	8.33 (2.25 to 30.92)	0.08 (0.02 to 0.32)	0.91	92.59 (75.71 to 99.09)
Damms 2008 [ELISA]	50	0.79 (0.60 to 0.88)	1.00 (0.93 to 1.00)	4.71 (2.96 to 7.50)	0	0.83	21.43 (13.22 to 31.74)
Damms 2008 [POCT]	50	0.55 (0.36 to 0.74)	0.96 (0.87 to 1.00)	4.51 (2.70 to 7.54)	0.14 (0.04 to 0.51)	0.82	21.43 (13.22 to 31.74)
Diamanti 2010	100	0.82 (0.75 to 0.88)	1.0 (0.93 to 1.0)	3.08 (2.24 to 4.22)	0 0	0.87	59.39 (52.18 to 66.31)
Diamanti 2010	160	0.88 (0.81 to 0.93)	1.00 (0.94 to 1.00)	5.00 (3.23 to 7.75)	0 0	0.92	59.39 (52.18 to 66.31)
Fagerberg 2005	50	0.96 (0.77 to 1.00)	0.93 (0.66 to 1.00)	13.36 (2.02 to 88.54)	0.05 (0.01 to 0.33)	0.94	61.11 (43.46 to 76.86)
Henderson 2012	>50	0.62 (0.53 to 0.70)	0.96 (0.85 to 0.99)	1.8 (.15 to 2.1)	0.05 (0.01 to 0.20)	0.70	47.89 (40.61 to 55.25)
Henderson 2012	>100	0.68 (0.59 to 0.76)	0.95 (0.86 to 0.99)	2.3 (1.8 to 3.0)	0.06 (0.02 to 0.17)	0.77	47.89 (40.61 to 55.25)
Henderson 2012	>200	0.77 (0.67 to 0.84)	0.92 (0.84 to 0.97)	3.6 (2.5 to 5.0)	0.09 (0.04 to 0.20)	0.83	47.89 (40.61 to 55.25)
Henderson 2012	>300	0.83 (0.74 to 0.90)	0.89 (0.81 to 0.95)	5.2 (3.3 to 8.0)	0.13 (0.07 to 0.24)	0.86	47.89 (40.61 to 55.25)
Henderson 2012	>800	0.93 (0.84 to 0.98)	0.79 (0.71 to 0.86)	14.5 (6.1 to 34.4)	0.29 (0.21 to 0.41)	0.84	47.89 (40.61 to 55.25)
Licata 2012	150	0.82 (0.74 to 0.88)	0.84 (0.78 to 0.88)	6.40 (4.35 to 9.44)	0.28 (0.21 to 0.37)	0.83	41.04 (35.81 to 46.43)
Limburg 2000	100	0.63 (0.46 to 0.78)	0.93 (0.85 to 98)	4.79 (2.89 to 7.93)	0.21 (0.09 to 0.47)	0.83	26.36 (18.42 to 35.62)
Sidler 2008	50	0.76 (0.60 to 0.88)	1.00 (0.83 to 1.00)	3.00 (1.81 to 4.98)	0.00	0.84	50.82 (37.70 to 63.86)
Sidler 2008	100	0.97 (0.82 to 1.00)	0.91 (0.75 to 0.98)	27.10 (3.93 to 186.78)	0.10 (0.03 to 0.29)	0.93	50.82 (37.70 to 63.86)
Tomas 2007	50	0.94 (0.70 to 1.00)	1.00 (0.74 to 1.00)	13.00 (1.98 to 85.46)	0	0.96	53.57 (33.87 to 72.49)
Tomas 2007	100	0.93 (0.68 to 1.00)	0.92 (0.63 to 1.00)	12.13 (1.98 to 80.15)	0.07 (0.01 to 0.48)	0.93	53.57 (33.87 to 72.49)
Tomas 2007	150	1.00 (0.75 to 1.00)	0.87 (0.60 to 0.99)		0.13 (0.04 to 0.48)	0.93	53.57 (33.87 to 72.49)
Tomas 2007	200	1.00 (0.74 to 1.00)	0.81 (0.54 to 0.96)		0.20 (0.07 to 0.55)	0.89	53.57 (33.87 to 72.49)
van de Vijver 2012	50	0.68 (0.55 to 0.79)	1.00 (0.94 to 1.00)	3.8 (2.6 to 5.5)	0	0.83	35.9 (27.24 to 45.29)
van de Vijver 2012 [excluding gastrointestinal]	50	0.78 (0.64 to 0.88)	1.00 (0.93 to 1.00)	5.17 (3.11 to 8.59)	0	0.88	40.38 (30.87 to 50.46)

At a cut-off of 50 µg/g, the overall results pooled for IBD versus IBS, show very high sensitivity (99%: 95% CI 95-100%) (figure 13) but moderate specificity (74%), probably because there are

organic conditions with raised calprotectin in the non-IBD group. There is moderate heterogeneity for sensitivity but high for specificity (figure 14).

At a cut-off of 100 µg/g, sensitivity falls to 94% (95% CI 86 – 98%) but specificity improves to 82% (95% CI 67 to 91%).

Henderson et al report results from a relatively large group of children, by linking referrals to the regional paediatric gastroenterology service (5,600) with laboratory calprotectin results (4,155 results) and endoscopy records, to create a cohort of 190 children investigated for possible IBD, who all had calprotectin and full endoscopy records.[26] 91 were shown to have IBD, of whom 62 had CD, 21 UC and the other 8 unclassified IBD. The pre-test probability of IBD was 0.48. They classed calprotectin results as;

- Normal <50 µg/g
- Possible inflammation 51-100 µg/g
- GI inflammation 101-200 µg/g
- Active GI inflammation >200 µg/g

and comment that in practice, they find the cut-off of 200 µg/g as being the most useful for likely diagnosis of IBD.

They provide results for four thresholds for positivity as follows in Table 20.

Table 20. Measures of diagnostic accuracy for increasing faecal calprotectin levels in Henderson 2012

FC cut-off (µg/g)	Se	Sp	NPV
>50	0.98	0.44	0.96
>100	0.97	0.50	0.95
>200	0.93	0.74	0.92
> 300	0.89	0.83	0.83

These nicely show the trade-off between sensitivity and specificity.

(They also provide data for a cut-off of >800 µg/g but sensitivity drops too much for that to be useful.)

Henderson et al also provide data that shows the different mix of non-IBD conditions in children. The non-IBD conditions included IBS (about a third of cases), non-specific colitis, post-infectious enteropathy, cow's milk or wheat intolerance, pinworms, and allergic enteropathy.

Ashorn et al included three serological markers, all of which reflect immune response to commensal intestinal bacteria;[79]

- ASCA – anti-Saccharomyces cerevisiae antibodies
- OmpW, antibodies against an outer membrane protein of Bacteroides caccae
- Antibodies against *I2* from *Pseudomonas fluorescens*.

The sensitivity of these was much poorer than that of calprotectin overall in IBD, but higher in CD (67%) than in UC (14%).

Canani et al also examined the use of serum markers in children.[60] They found sensitivities of 41% for CRP, 52% for ESR, 78% for ASCA/pANCA, and 93% for calprotectin.

Fagerberg reported data on faecal occult blood, which was less useful than calprotectin (cut-off 50 µg/g).[81] Se, Sp, PPV and NPV for calprotectin were 95%, 93%, 95% and 93%, and for faecal occult blood, 58%, 91%, 92% and 56% respectively. ESR and CRP had poor sensitivities of 41% and 36%, and NPVs of 52% and 50%. All children with IBD had calprotectin levels >50 µg/g. The 95% sensitivity arose because it was expressed in terms of inflammation, not IBD, and one child had a normal calprotectin (15 µg/g) but non-specific proctitis. One child with no mucosal inflammation identified had a calprotectin of 65 µg/g.

Fagerberg et al note that 60% of the children in their study had colonic inflammation, which they consider to be typical of paediatric groups being investigated – a contrast from adult groups with their much lower prevalence of IBD, due to the commonness of IBS.

Shaoul et al (not included in our meta-analysis) provide a report on calprotectin levels in children (age range 8 to 17 years) with untreated CD (a case series with no non-IBD comparison group, hence an exclusion for our purposes).[83] The title of their paper is "limitations of fecal calprotectin" but this appears to be chosen for two reasons. Firstly, they note a lack of correlation with PCDAI. However, this may reflect the limitations of clinical activity scores rather than of calprotectin. Secondly, in their group of 60 children with CD, three had normal calprotectin levels. Two of these had "minimal" findings on endoscopy, and the other had moderate changes. Interestingly, two of the children had internal fistulae at diagnosis. They also report CRP and ESR results: 8 of 10 patients with normal CRP had raised calprotectin, as did 9 of 10 with normal ESR.

Sidler et al compared faecal calprotectin with faecal S100A12, CRP and ESR.[82] S100A12 is a protein from the same S100 family as calprotectin (which is a complex of S100A8 and S100A9). S100A12 performed better than calprotectin because of specificity. Sensitivities were similar at 100% for calprotectin and 97% for S100A12, but specificities were 67% and 97% respectively. NPVs were

97% for S100A12 and 100% for calprotectin (data from table - text says NPV for calprotectin was 95%). ESR had 74% sensitivity and CRP 81%. Serum S100A12 had sensitivity of only 22%. The 67% Sp for calprotectin is considerably lower than in other studies such as Fagerberg (93%).

All but one of the 31 patients with IBD, had CD. The low specificity of calprotectin was due to raised levels in some children without IBD. These had various conditions, including *Helicobacter pylori* infection, a duodenal ulcer, and reflux oesophagitis, but the paper does not say which had the raised calprotectin.

Bremner et al (not included in meta-analysis) report a study of calprotectin in children, but most were not newly diagnosed and so it was an exclusion for our purposes.[84] However they noted that some children without bowel inflammation had raised (>50 μg/g) calprotectin. Three had functional constipation on laxative treatment, and one had normal findings but a family history of IBD. The latter raises the possibility that calprotectin may be raised before there is clinical evidence of IBD. As in adults, calprotectin is not raised in eosinophilic, lymphocytic or non-specific colitis.[85]

In Figure 12, specificity is rather more variable than sensitivity, and confidence intervals also vary. The precision of both depends on patient numbers. For example, in studies having a higher proportion of patients with disease than without, estimates of sensitivity will be more precise than those of specificity. This is best illustrated by the Diamanti study.

Study	TP	FP	FN	TN	Cut-off level	Make of test	Age	Primary of secondary care	ELISA or POCT	Sensitivity (95% CI)	Specificity (95% CI)
Fagerberg 2005	21	1	1	13	50.0	Calprest	Paediatric	Secondary	ELISA	0.95 [0.77, 1.00]	0.93 [0.66, 1.00]
Tomas 2007a	15	1	0	12	50.0	Calprest	Paediatric	Secondary	ELISA	1.00 [0.78, 1.00]	0.92 [0.64, 1.00]
Sidler 2008	31	10	0	20	50.0	PhiCal	Paediatric	Secondary	ELISA	1.00 [0.89, 1.00]	0.67 [0.47, 0.83]
Damms 2008b	16	13	2	53	50.0	Prevista	Adult	Secondary	POCT	0.89 [0.65, 0.99]	0.80 [0.69, 0.89]
Damms 2008a	18	14	0	52	50.0	EK-CAL	Adult	Secondary	ELISA	1.00 [0.81, 1.00]	0.79 [0.67, 0.88]
Van de Vijver 2012a	42	20	0	55	50.0	CALPRO	Paediatric	Secondary	ELISA	1.00 [0.92, 1.00]	0.73 [0.62, 0.83]
Henderson 2012a	89	55	2	44	50.0	PhiCal	Paediatric	Secondary	ELISA	0.98 [0.92, 1.00]	0.44 [0.34, 0.55]
Canani 2006	25	2	2	16	93.5	Calprest	Paediatric	Secondary	ELISA	0.93 [0.76, 0.99]	0.89 [0.65, 0.99]
Tomas 2007b	14	1	1	12	100.0	Calprest	Paediatric	Secondary	ELISA	0.93 [0.68, 1.00]	0.92 [0.64, 1.00]
Limburg 2000	24	14	5	67	100.0	PhiCal	Adult	Secondary	ELISA	0.83 [0.64, 0.94]	0.83 [0.73, 0.90]
Sidler 2008b	28	1	3	29	100.0	PhiCal	Paediatric	Secondary	ELISA	0.90 [0.74, 0.98]	0.97 [0.83, 1.00]
Ashorn 2009	39	1	5	10	100.0	PhiCal	Paediatric	Secondary	ELISA	0.89 [0.75, 0.96]	0.91 [0.59, 1.00]
Diamanti 2010a	117	26	0	54	100.0	Calprest	Paediatric	Secondary	ELISA	1.00 [0.97, 1.00]	0.68 [0.56, 0.78]
Henderson 2012b	88	41	3	58	100.0	PhiCal	Paediatric	Secondary	ELISA	0.97 [0.91, 0.99]	0.59 [0.48, 0.68]
Tomas 2007c	13	0	2	13	150.0	Calprest	Paediatric	Secondary	ELISA	0.87 [0.60, 0.98]	1.00 [0.75, 1.00]
Licata 2012	107	24	35	180	150.0	Calprest	Adult	Secondary	ELISA	0.75 [0.67, 0.82]	0.88 [0.83, 0.92]
Diamanti 2010b	117	16	0	64	160.0	Calprest	Paediatric	Secondary	ELISA	1.00 [0.97, 1.00]	0.80 [0.70, 0.88]
Tomas 2007d	12	0	3	13	200.0	Calprest	Paediatric	Secondary	ELISA	0.80 [0.52, 0.96]	1.00 [0.75, 1.00]
Henderson 2012c	85	26	6	73	200.0	PhiCal	Paediatric	Secondary	ELISA	0.93 [0.86, 0.98]	0.74 [0.64, 0.82]
Henderson 2012d	81	17	10	82	300.0	PhiCal	Paediatric	Secondary	ELISA	0.89 [0.81, 0.95]	0.83 [0.74, 0.90]
Henderson 2012e	66	5	25	94	800.0	PhiCal	Paediatric	Secondary	ELISA	0.73 [0.62, 0.81]	0.95 [0.89, 0.98]

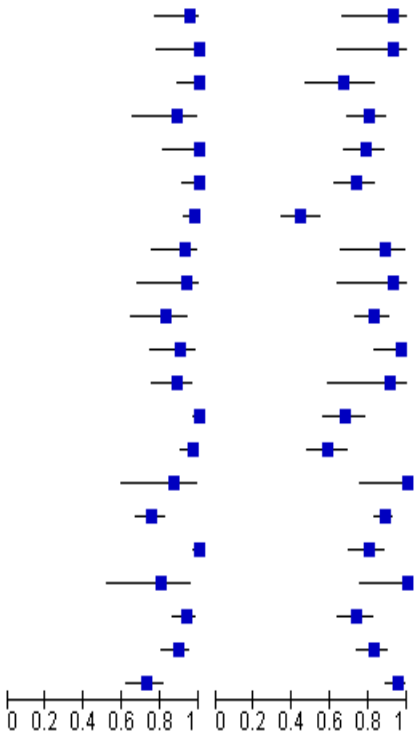


Figure 12. IBD vs non-IBD

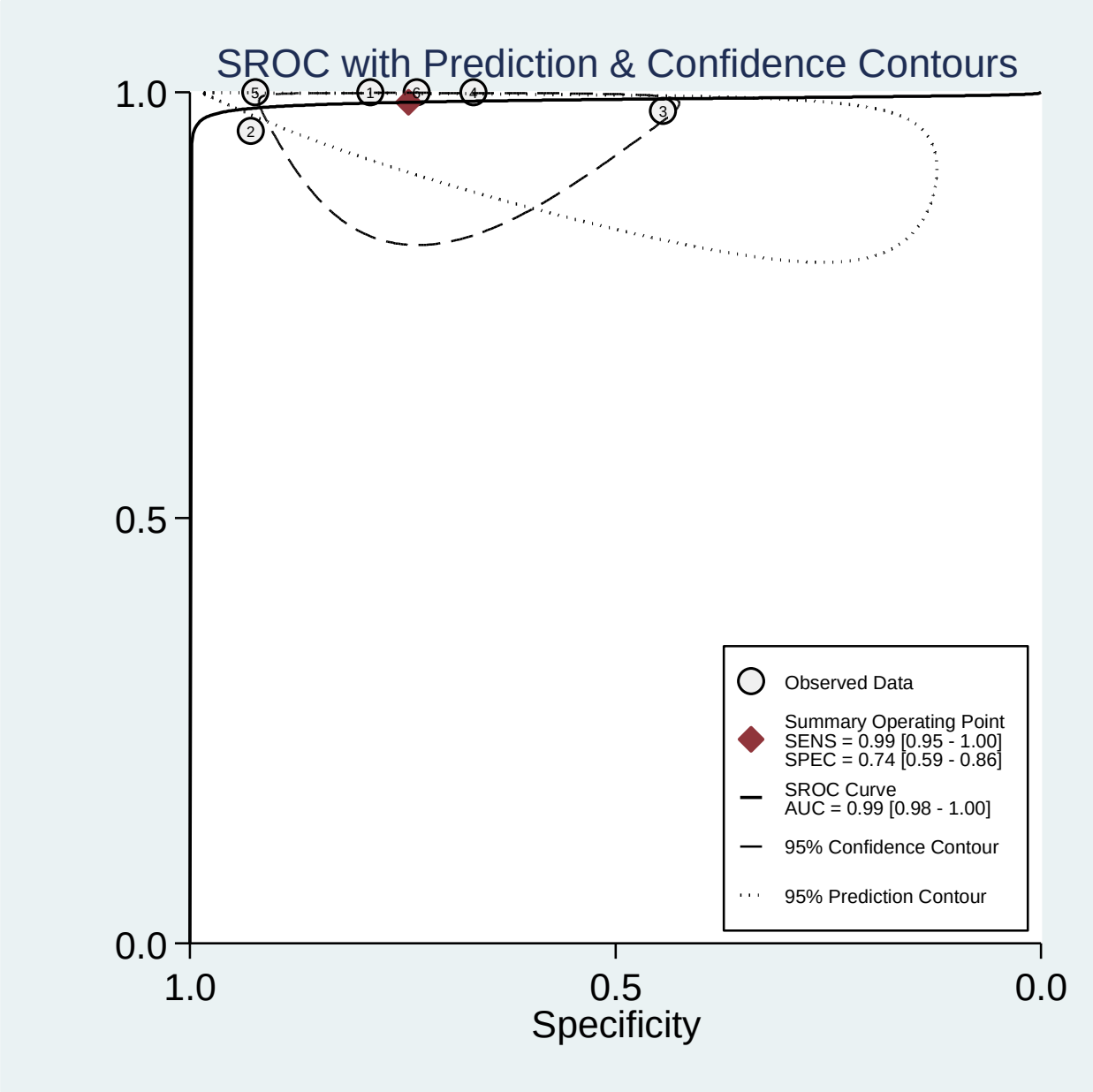


Figure 13. Summary receiver-operating characteristic (sROC) curve for fecal calprotectin in the diagnosis of bowel diseases IBD versus non-IBD at a cut-off level of 50 µg/g

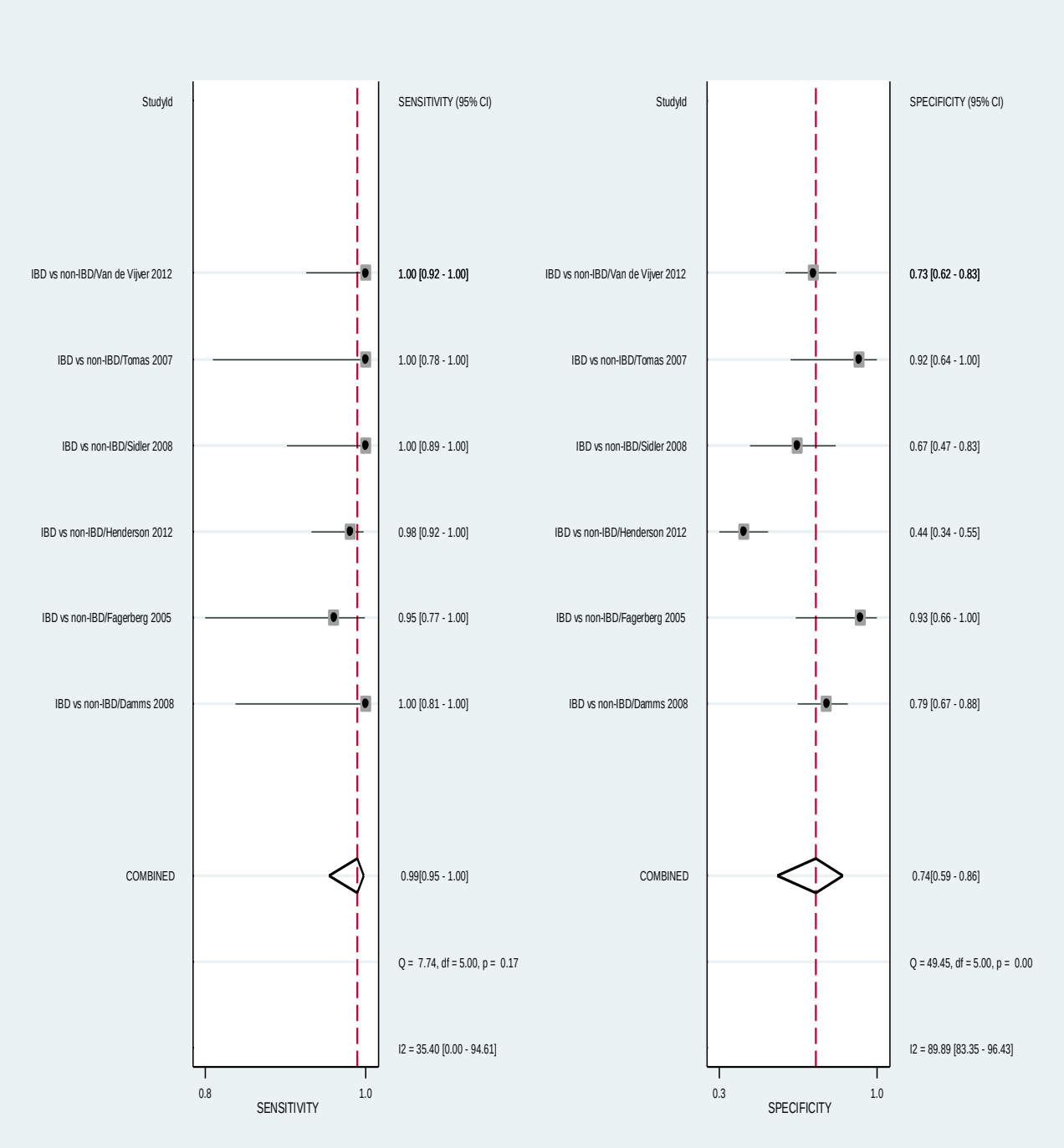


Figure 14. Forest plots of pooled sensitivity and specificity of fecal calprotectin in the diagnosis of bowel diseases IBD versus non-IBD at a cut-off level of 50 µg/g

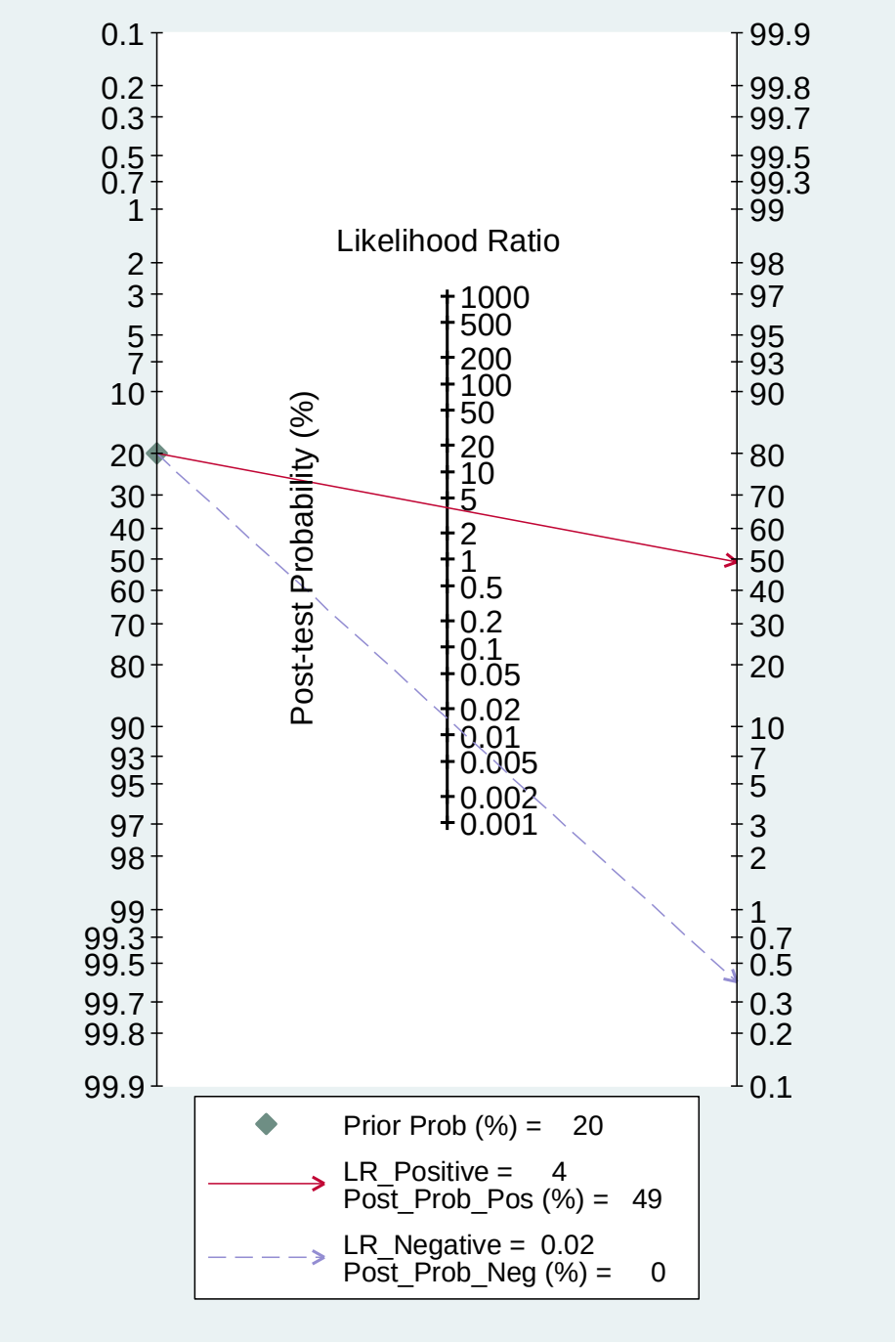


Figure 15. The use of the Fagan's nomogram (a straight line through the pre-test probability of 20% and the LR− of 0.20 yields a post-test probability of about 2%). [IBD versus non-IBD at a cut-off level of 50 µg/g].

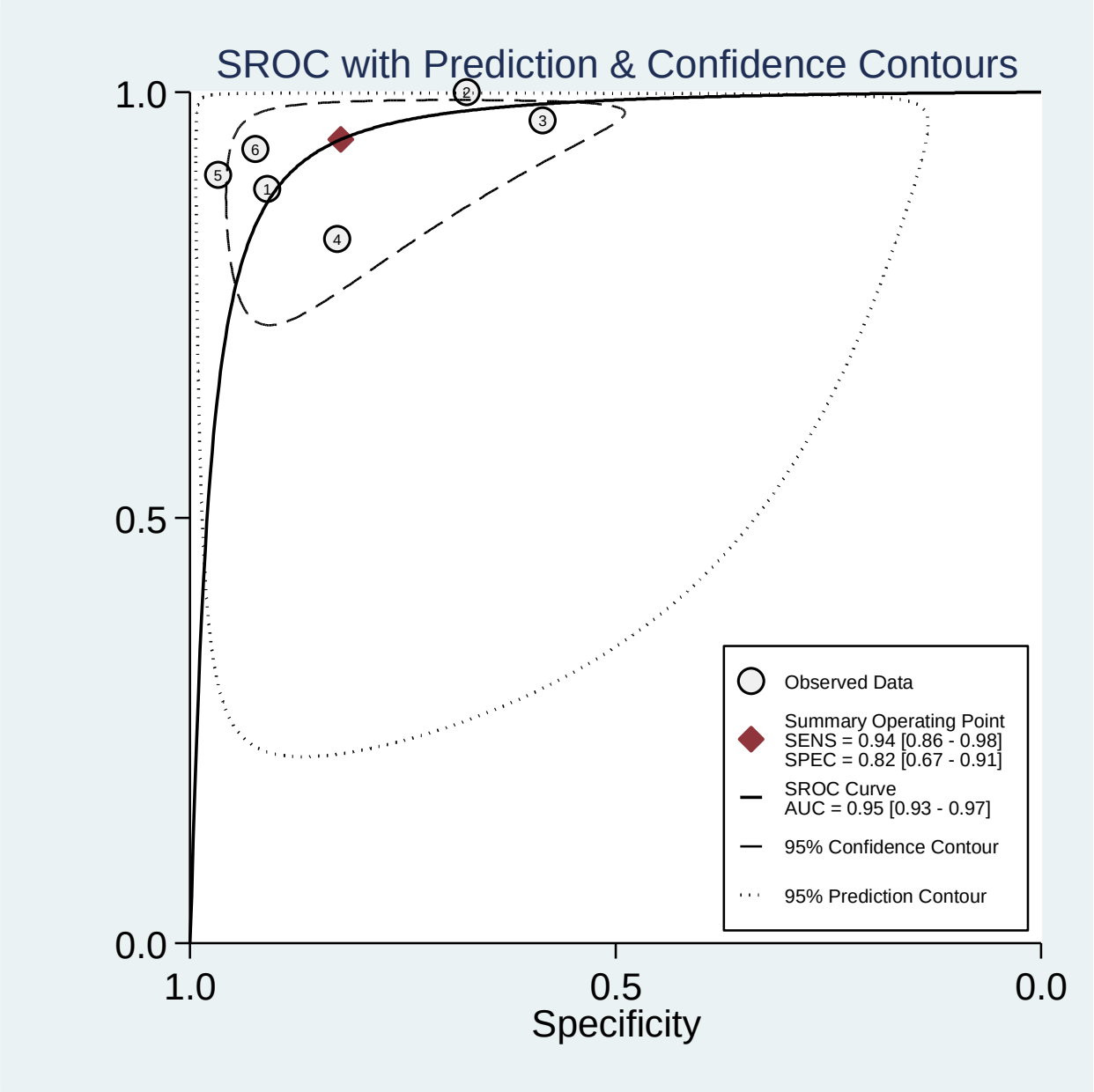


Figure 16. Summary receiver-operating characteristic (sROC) curve for fecal calprotectin in the diagnosis of bowel diseases IBD versus non-IBD at a cut-off level of 100 µg/g

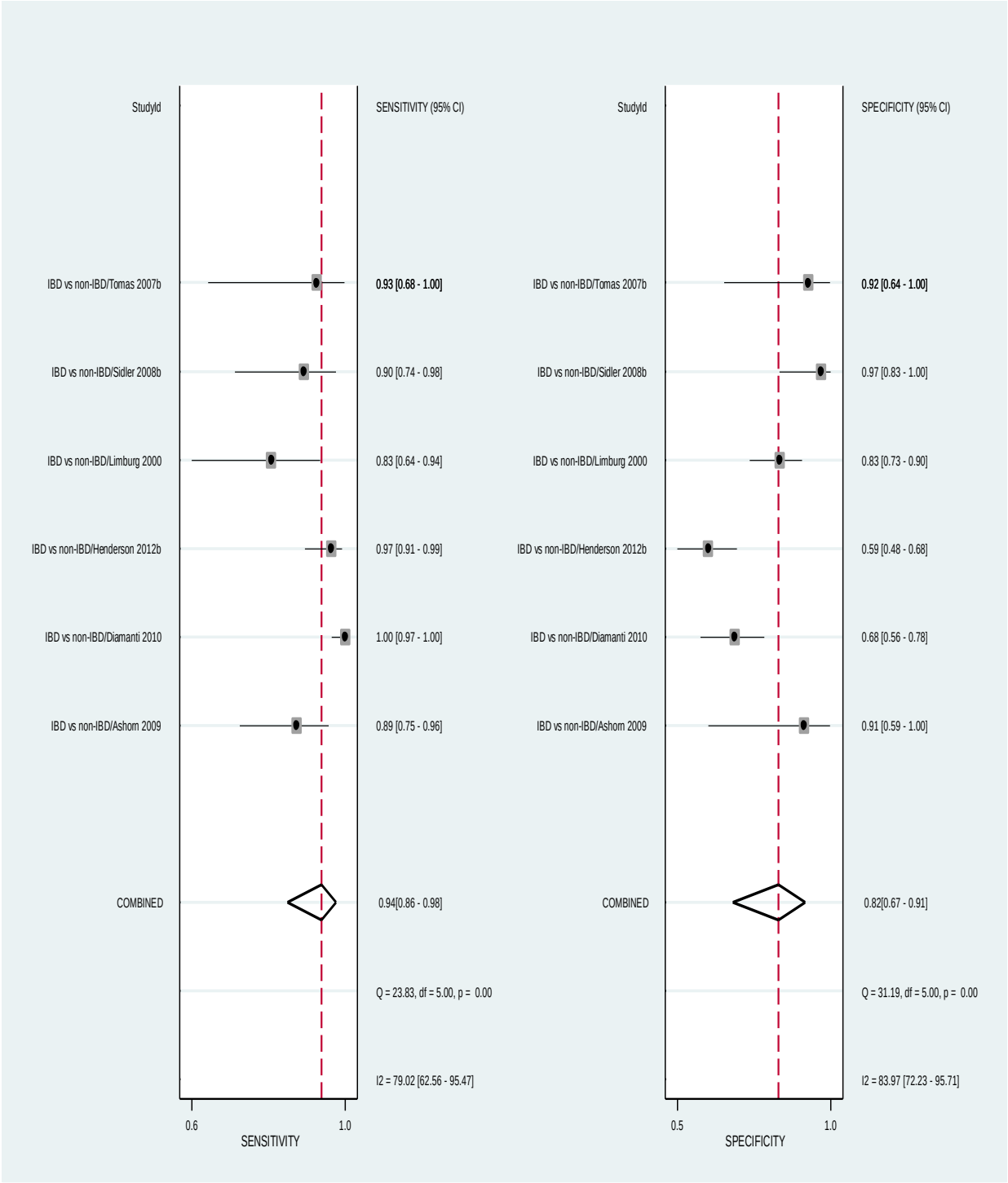


Figure 17. Forest plots of pooled sensitivity and specificity of fecal calprotectin in the diagnosis of bowel diseases IBD versus non-IBD at a cut-off level of 100 µg/g

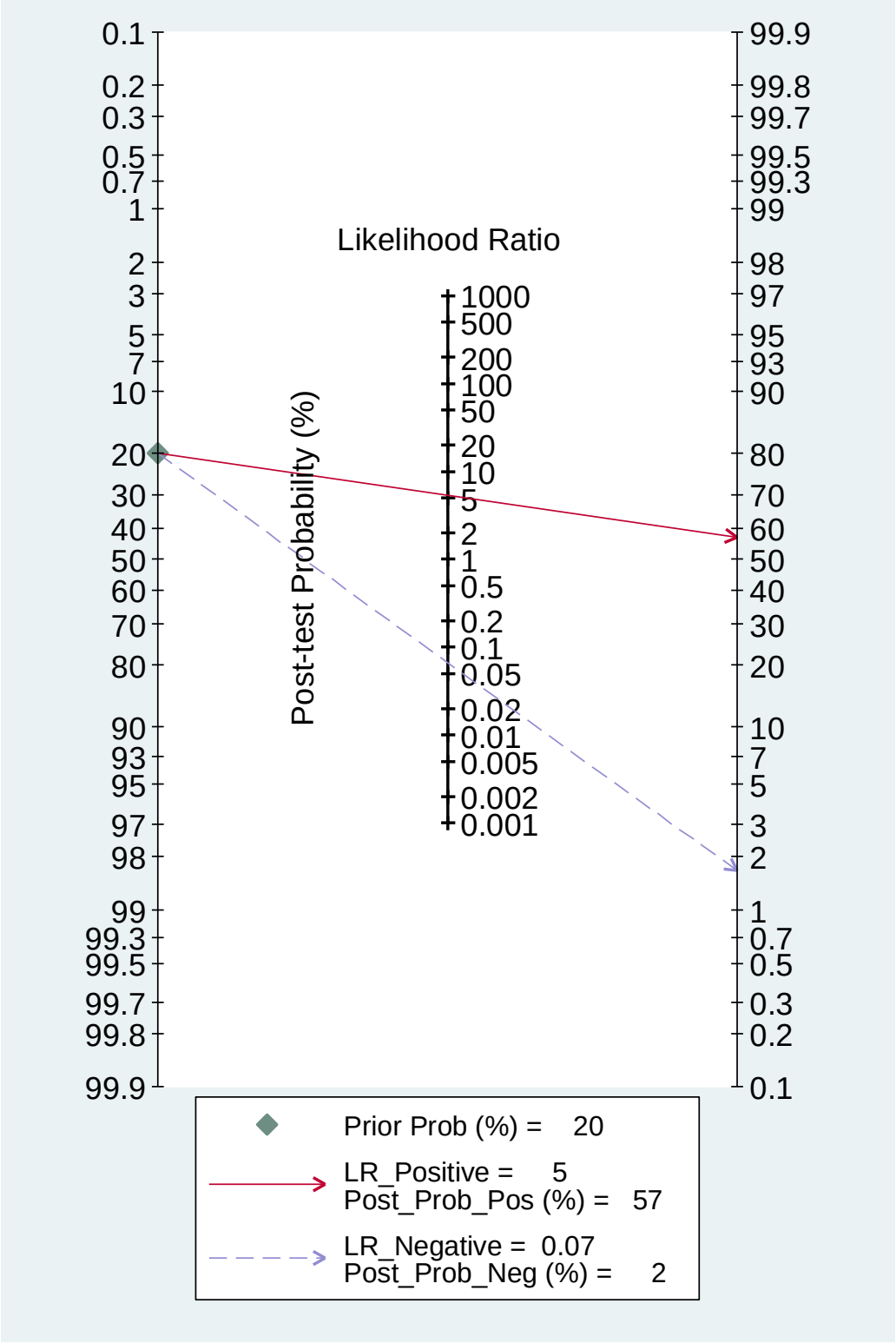


Figure 18. The use of the Fagan's nomogram (a straight line through the pre-test probability of 20% and the LR− of 0.20 yields a post-test probability of about 2%). [IBD versus non-IBD at a cut-off level of 100 µg/g].

Table 21. Diagnostic Odds Ratios: IBD vs non-IBD studies

Number of Patients	Number of Studies	Cut-off level	DOR (95% CI)
531	6	50	246 (44 to 1376)
45	1	93.5	100 (10.2 to 1250.0)
656	6	100	79 (31 to 202)
389	2	150	5.1 (3.8 to 6.9)
197	1	160	114 (7.2 to 1804)
233	2	200	8.5 (4.5 to 15.8)
190	1	300	39.1 (15.8 to 99.9)
190	1	800	49.6 (17.2 to 169.8)

Summary IBD vs non-IBD

In these mostly paediatric studies, the overall results pooled for IBD versus IBS, show very high sensitivity (99%: 95% CI 95-100%) (Figure 13) but moderate specificity (74%) at a cut-off of 50 µg/g. At a cut-off of 100 µg/g, sensitivity falls to 94% (95% CI 86 – 98%) but specificity improves to 82% (95% CI 67 to 91%).

Calprotectin is therefore a valuable test in children with suspected IBD, and will allow most with non-IBD conditions to avoid invasive investigations, in particular colonoscopy.

2.8 Studies of calprotectin: organic versus non-organic bowel disease

Table 22. Outline of studies comparing organic vs non-organic bowel disease

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
Burri 2013[86]	405	Patients undergoing endoscopy of the GI tract for abdominal discomfort	Department of Gastroenterology of the University Hospital Basel, Switzerland.	To compare three different assays in their ability to identify patients with organic intestinal disease	Endoscopy, esophagogastroduodenoscopy and histology	Younger than 18 years
Dolwani 2004[42]	63	Consecutive patients undergoing small bowel BaFT examination	Gastroenterology outpatient clinic, University of Wales Hospital.	To compare the utility of a single FC estimation to barium follow through in exclusion of intestinal inflammation	Rigid sigmoidoscopy and stool cultures	Known malignancy, on NSAIDs, coeliac diseases etc
Garcia 2006[54]	190	Consecutive individuals who underwent colonoscopy for medical indications	Hospital Universitario Reina Sofia Córdoba, Spain	To assess the usefulness of FC to predict the presence of pathological colonoscopy	Colonoscopy, clinical criteria, endoscopic and histologic findings.	Severe cardiopulmonary disease, kidney or liver

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
						disease, celiac disease, known malignancy
Kok 2012[33]	382	Patients consulting their GPs for persistent lower-abdominal complaints	Data from the CEDAR study in 170 general practices in 2 regions of the Netherlands	To quantify the diagnostic accuracy of 3 biomarker tests for the inclusion or exclusion of OBD	Endoscopy, and biopsies if required.	<18 years old, previously diagnosed with OBD, or positive on triple faeces test
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
Manz 2012[88]	538	Patients undergoing endoscopy of the GI tract for abdominal discomfort	Department of Gastroenterology of the University Hospital Basel in Switzerland.	To prospectively investigate the value of FC as a biological marker for the diagnosis of intestinal organic disease in symptomatic patients	Endoscopy and biopsies as decided by the endoscopist.	Younger than 18 years old.
Shitrit 2007[57]	69	Patients referred to the Department of Gastroenterology for colonoscopic examination of various indications, including screening.	Department of Gastoenterology, Shaare-Zedek Medical Center, Israel	To assess the predictive value of FC in organic colonic disease.	Colonoscopy and histopathology.	Intake of NSAIDs and/or antibiotics during the previous three months , concomitant serious illness
Tibble 2002[43]	602	Patients referred to a gastroenterology outpatient department by GPs.	Gastroenterology outpatient department of a teaching hospital in South London.	To determine if the use of FC and intestinal permeability are useful in differentiating between patients with organic and nonorganic disease.	One or more invasive diagnostic imaging procedures,	Previously diagnosis of IBD, colore

Study	Number of patients	Recruits	Setting	Aim	Reference test	Exclusions
					appropriate to their symptoms.	ctal carcinoma, and other serious diseases
Tomas 2007[61]	43	Referred by GPs; all patients had clinical symptoms suggestive of organic intestinal disease or IBS that had not responded to therapy	Patients referred to the Pediatric GI Unit of a hospital in Mallorca, Spain.	To evaluate FC in paediatric patients with signs and symptoms suggestive of IBD	Clinical criteria, laboratory, image and endoscopic test results	Not reported
Turvill 2012[40]	630	New patient referrals from primary care, aged 16–60 years, with intestinal symptoms.	The Department of Gastroenterology, York Hospital.	To determine the NPV of a normal FC in excluding organic intestinal disease in patients with intestinal symptoms	Colonoscopy,supportive histology, barium meal, CT enterography and capule endoscopy.	Patients with fast track colorectal symptoms.

Table 23. QUADAS quality assessment of studies comparing organic vs non-organic bowel disease

	Burri 2013	**Dolwani 2004**	**Garcia 2006**	**Kok 2012**	[redacted]	**Manz 2012**	**Shitrit 2007**	**Tibble 2002**	**Tomas 2007**	**Turvill 2012**
Spectrum?	Yes	Yes	Yes	Yes	[redacted]	Yes	Yes	Yes	Unclear	Yes
Reference standard?	Yes	Yes	Yes	Yes	[redacted]	Yes	Yes	Yes	Yes	Unclear
Acceptable delay?	Unclear	Yes	Yes	Yes	[redacted]	Unclear	Yes	Yes	Unclear	Unclear
Whole sample verified?	Unclear	Yes	Yes	Yes	[redacted]	Yes	Yes	Yes	Yes	No
Same reference standard	No	Yes	Yes	Yes	[redacted]	No	Yes	Yes	Unclear	Yes
Newly diagnosed?	Yes	Yes	Unclear	Yes	[redacted]	Yes	Unclear	Yes	Yes	Yes
Blinded reference testing?	Yes	Yes	Unclear	Yes	[redacted]	Yes	Yes	Unclear	Unclear	Unclear
Index results blinded?	Yes	Yes	Unclear	Yes	[redacted]	No	Unclear	Unclear	Unclear	Unclear
Same clinical data	Yes	Unclear	Yes	Yes	[redacted]	Yes	Yes	Yes	Yes	Yes
Intermediate results reported?	Yes	Yes	Yes	Yes	[redacted]	Yes	Yes	Yes	Unclear	Unclear
Withdrawals explained?	Yes	Yes	Unclear	Yes	[redacted]	Yes	Yes	Yes	Yes	Unclear

Table 24. Table of results comparing organic vs non-organic bowel disease

Study	Cut-off value (µg/g)	PPV (95% CI)	NPV (95% CI)	PLR (95% CI)	NLR (95% CI)	Accuracy	Disease Prevalence % (95% CI)
Burri 2013	50	0.90 (0.83 to 0.94)	0.89 (0.85 to 0.92)	15.78 (9.23 to 27.00)	0.23 (0.17 to 0.31)	0.89	35.31 (30.65 to 40.18)
Burri 2013	22.4	0.84 (0.75to 0.91)	0.84 (0.78 to 0.87)	9.67 (5.93 to 15.77)	0.36 (0.28 to 0.46)	0.84	35.18 (30.25 to 40.35)
Burri 2013	50	0.93 (0.84 to 0.98)	0.76 (0.71 to 0.81)	25.33 (9.40 to 68.30	0.58 (0.49 to 0.67	0.79	35.18 (30.25 to 40.35)
Dolwani 2004	60	0.60 (0.39 to 0.79)	1.00 (0.91 to 1.00)	4.80 (2.77 to 8.33)	0	0.84	23.81 (13.98 to 36.21)
Garcia 2006	217	0.74 (0.63 to 0.83)	0.90 (0.82 to 0.95)	4.52 (3.06 to 6.66)	0.19 (0.11 to 0.32)	0.83	38.42 (31.47 to 45.74)
Kok 2012	50	0.24 (0.19 to 0.31)	0.92 (0.87 to 0.95)	1.7 (1.4-2.0)	0.5 (0.3-0.7)	0.58	16.23 (12.68 to 20.32)
Kok 2012	50	0.22 (0.17 to 0.28)	0.93 (0.88 to 0.96)	1.5 (1.3-1.7)	0.4 (0.2-0.7)	0.51	16.23 (12.68 to 20.32)
Kok 2012	50	0.32 (0.26 to 0.39)	0.81 (0.74 to 0.86)	1.4 (1.1 -1.7)	0.7 (0.5 to 0.9)	0.56	25.92 (21.59 to 30.62)
Kok 2012	50	0.33 (0.27 to 0.39)	0.84 (0.77 to 0.89)	1.4 (1.2-1.6)	0.6 (0.4-0.8)	0.54	25.92 (21.59 to 30.62)
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Manz 2012	50	0.87 (0.81 to 0.92)	0.84 (0.80 to 0.88)	10.36 (6.93 to 15.50)	0.29 (0.23 to 0.36)	0.85	39.41 (35.25 to 43.68)
Manz 2012	10	0.55 (0.50 to 0.60)	0.93 (0.88 to 0.96)	1.88 (1.68 to 2.10)	0.12 (0.07 to 0.21)	0.67	39.41 (35.25 to 43.68)
Manz 2012	48	0.68	0.75	3.23	0.5		
Manz 2012	50	0.93	0.82	10.6	0.17		
Shitrit 2007	150	0.75 (0.55 to 0.89)	0.83 (0.68 to 0.93)	4.39 (2.16 to 8.91)	0.30 (0.16 to 0.58)	0.80	40.58 (28.91 to 53.08)
Tibble 2002	10 mg/L = 50 µg/g	0.77 (0.72 to 0.81)	0.90 (0.86 to 0.93)	4.25 (3.44 to 5.25)	0.14 (0.10 to 0.20)	0.83	43.69 (39.68 to 47.76)
Tomas 2007	50	0.96 (0.80 to 1.00)	0.71 (0.44 to 0.90)	10.83 (1.64 to 71.7)	0.18 (0.08 to 0.41)	0.86	69.77 (53.90 to 82.80)
Tomas 2007	100	0.96 (0.78 to 1.00)	0.60 (0.36 to 0.81)	9.53 (1.43 to 63.45)	0.29 (0.16 to 0.53)	0.79	69.77 (53.90 to 82.80)
Tomas 2007	150	1.00 (0.80 to 1.00)	0.50 (0.30 to 0.70)		0.43 (0.29 to 0.65)	0.70	69.77 (53.90 to 82.80)
Tomas 2007	200	1.00 (0.75 to 1.00)	0.43 (0.25 to 0.63)		0.57 (0.41 to 0.77)	0.60	69.77 (53.90 to 82.80)
Tomas 2007	50	0.94 (0.70 to 1.00)	1.00 (0.74 to 1.00)	13.00 (1.98 to 85.46)	0	0.96	53.57 (33.87 to 72.49)
Tomas	100	0.93 (0.68 to 1.00)	0.92 (0.63 to 1.00)	12.13 (1.98 to 80.15)	0.07 (0.01 to 0.48)	0.93	53.57 (33.87 to 72.49)

Study	Cut-off value (µg/g)	PPV (95% CI)	NPV (95% CI)	PLR (95% CI)	NLR (95% CI)	Accuracy	Disease Prevalence % (95% CI)
2007							
Tomas 2007	150	1.00 (0.75 to 1.00)	0.87 (0.60 to 0.99)		0.13 (0.04 to 0.48)	0.93	53.57 (33.87 to 72.49)
Tomas 2007	200	1.00 (0.74 to 1.00)	0.81 (0.54 to 0.96)		0.20 (0.07 to 0.55)	0.89	53.57 (33.87 to 72.49)
Turvill 2012	<50	0.77 (0.68 to 0.84)	0.96 (0.95 to 0.98)	16.11 (11.05 to 23.48)	0.17 (0.11 to 0.27)	0.93	17.30 (14.43 to 20.49)
Turvill 2012	< 60	0.81	0.96				
Turvill 2012	<75	0.86	0.93				
Turvill 2012	< 100	0.91	0.91				

Figure 19 has been updated. Please refer to the Diagnostics Assessment Report Addendum

Figure 19. Organic vs non-organic bowel disease

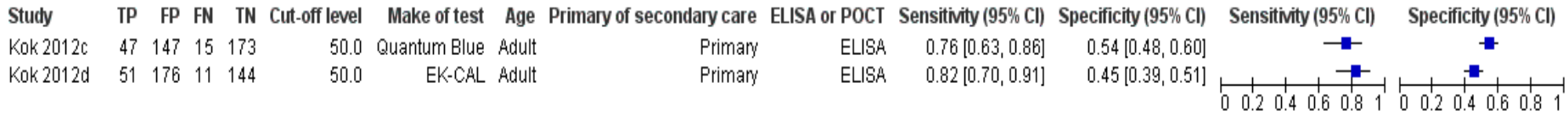


Figure 20. Organic (excluding adenomas ≤ 1 cm) vs non-organic bowel disease

Table 25. organic vs non-organic bowel disease

Number of Patients	Cut-off level	Number of Studies	DOR (95% CI)
538	10	1	15.3 (8.3 to 30.3)
405	22.4	1	26.6 (13.7 to 53.0)
3005	50	7	33 (13 to 81)
638	60	1	46.5 (2.9 to 743.6)
43	100	1	33 (3.5 to 1472.1)
112	150	2	2.8 (1.9 to 4.0)
43	200	1	12.2 (0.8 to 190.9)
190	217	1	24.3 (10.4 to 58.8)

The organic diseases that give rise to raised calprotectin include NSAID enteropathy, diverticular disease, polyps, and coeliac disease, but calprotectin can be normal in the presence of some of these, including polyps and diverticulosis.[40]

2.9 Ranges

It is worth noting that notwithstanding the generally good predictive value of calprotectin for differentiating IBD and IBS in adults, and IBD and non-IBD in children, the range of results can be wide, with some low levels in patients with IBD and raised levels in people with IBS.
The tables below give some examples of both full ranges and inter-quartile ranges. In some studies, the ranges do not overlap, in others they do. For example: in El-Badry 2010, the value of FC in patients with IBD ranged between 98 and 637 µg/g which does not overlap with the value of FC in patients with IBS (14 to 65 µg/g). In all other studies, the range of FC in patients with IBD overlapped with the range of FC in patients with IBS. In some studies, like Li 2006 and Schroder 2007, the range of FC level in patients with IBD was wide with the lowest value being 15 µg/g and the highest being 2574 µg/g.

Table 26. Adults: IBD vs IBS

	Range IBD	Range IBS	IQR IBD	IQR IBS
Carroccio 2003	180 to 400	10 to 210		
El-Badry 2010	98 to 637	14 to 65		
Kok 2012 ELISA			55-1200	21 to 99
Kok 2012 QB			64-300	30 to 69
Li 2006	23 to 2574	1 to 73	120-1118	6 to 27
Schroder 2007	15-2553	0 to 24		

The range of results in studies comparing IBD and non-IBD in children was similar to that found in studies comparing IBD and IBS in adults. In some studies (Canini 2006; Diamanti 2010; Sidler 2008), the ranges overlapped, in others they did not. . It should also be noted that in some patients with IBS,

FC levels were high, considerably more than the manufacturer's cut-off levels (Canani 2006; Diamanti 2010; Sidler 2008).

Table 27. Children: IBD vs non-IBD

	Range IBD	Range non-IBD	IQR IBD.	IQR non-IBD
Ashorn 2009	90 to 2,250 (CD); 105 to 900 (IC) 5 to 2600 (UC)	0 to 90		
Canani 2006	150 to 800 (CD) 200 to 1100 (UC)	0 to 160		
Diamanti 2010	162 to 9500	15 to 400		
Fagerberg 2005	213 to 440	7 to 28		
Sidler 2008	52 to 12000	19 to 201		
Tomas 2007			322 to 2967	36 to 193

2.10 Choice of test

Given that the focus of this review is in the performance of calprotectin tests for distinguishing patients who need to be referred from those who don't, we need to assess the comparative performance of different tests at levels representative of patients in that situation. These will consist of patients with IBS, most with normal or low calprotectins; and patients with IBD, some of who will have lowish calprotectins (50-200 µg/g), and some of whom will have florid inflammation and high levels. Given the low to high range of results, it may be unsafe to extrapolate from studies that compare different tests in groups of patients with much higher levels, such as when calprotectin is used to identify relapse, or to monitor treatment.

We therefore gave preference to studies in newly-presenting patients

Studies comparing FC tests: newly presenting patients.

Four studies (Damms 2008[53], Kok 2012[33], Otten 2008[65], Burri 2012[86]) reported studies where more than one type of commercial test kit was used shown in Table 28. Burri compared two different ELISAs and the other three studies used a rapid test and an ELISA in the same patients. All were in adult patients.

Otten 2008 tested the correlation between the POC test (Prevent ID CalDetect cut-off ≥15 µg/g compared to an ELISA (Phical CALPRO) cut-off >50 µg/g on 114 samples for distinguishing between IBD and IBS. The correlation between the two tests gave a Cohen's kappa = 0.69.

CalDetect had sensitivity of 1.00 (95% CI 0.85 to 1.00) and a specificity of 0.95 (0.88 to 0.98) compared to the PhiCal which gave sensitivity of 0.96 (0.78 to 1.00) and specificity of 0.87 (0.78 to 0.93). Therefore the rapid test at 15 µg/g was more sensitive and specific than the ELISA at 50 µg/g but neither difference was statistically significant.

Damms 2008 compared the Prevista POC and Bühlmann ELISA tests, both at a 50 µg/g cut-off, for detecting IBD, in 140 patients. The Bühlmann ELISA kit gave a sensitivity of 1.00 (0.82 to 1.00), specificity = 0.79 (0.67 to 0.88), area under the ROC (AUC) curve= 0.955. The Prevista rapid test sensitivity was 0.89 (0.65 to 0.98), and specificity was 0.80 (0.69 to 0.89), AUC= 0.896. So there were no significant differences in performance.

Kok 2012 compared Quantum Blue POC test with the vs EK-CAL ELISA in organic bowel disease (OBD) (which included all adenomas as OBD) vs non-OBD in 382 primary care patients, both at 50 µg/g cut-of levels. The agreement between the calprotectin POC and ELISA test was good [ICC 0.88 (0.85– 0.90), kappa = 0.66 (0.59–0.73)]

Note that the mean age was 60, so there was a higher prevalence of neoplasia, both benign and malignant, than would be expected in the age groups at which IBS was being separated from IBD. Of those investigated, 26% had OBD, of which 19% had colorectal cancer, and 54% had adenomas. Only 19% had IBD, with 7% had UC and 2% had CD.

The POC test had an AUC=0.66 (95% CI 0.60 to 0.72), similar to the ELISA AUC = 0.65 (95% CI 0.59 to 0.72). The sensitivity of the POC test was lower than the ELISA, with values of 0.64 (0.54-0.72) and 0.74 (0.64-0.82) respectively, but POC had a higher specificity 0.53 (0.48 - 0.59) vs 0.47 (0.41-0.53). As with the previous studies, these results overlap.

When small (1cm or less) adenomas were excluded from the OBD category, the performance of both tests improved, with increased AUCs, Sp and Se. The AUCs are still similar between POC and ELISA at 0.75 (0.67 to 0.72) and 0.73 (0.66 to 0.81) respectively. The POC test still has a lower Se = 0.76 (0.64 to 0.85) than the ELISA Se = 0.82 (0.70 to 0.91), but a higher specificity Sp = 0.54 (0.48 to 0.59) vs Sp = 0.46 (0.41 to 0.51);

Of the 19 patients with IBD, the POC test identified 15 and the ELISA 16, both at 50 µg/g.

Burri 2013 performed a post-hoc analysis of a prospective study (Manz 2012 – also included in this study) to compare two ELISA tests. The EK-CAL is monoclonal, and the PhiCAL is polyclonal.

The cut-off values used were >51 µg/g for EK-CAL (Buhlman ELISA) and >22.4 µg/g for PhiCal. These were optimal values cut-off values calculated from ROC analysis. The manufacturers' cut-off were both 50 µg/g.

Calprotectin concentrations measured by EK-CAL correlated better with PhiCal (rho=0.702, P <0.001) than with IBD-Scan (for lactoferrin) (rho=0.592, P <0.001). The mean (SD) of the difference) between the measurements of faecal calprotectin using EK-CAL and PhiCal was 30.9 (198.0) µg/g.

The AUC=0.918 for EK-CAL was significantly better than for PhiCal AUC = 0.842 p<0.001 (from text – figure 3 has slightly different AUCs).

EK-CAL ELISA at cut-off >51 µg/g had a Se = 0.78 (0.71 to 0.85), Sp = 0.95 (0.92 to 0.97) compared to PhiCal at cut-off >22.4 µg/g Se = 0.66 (0.57 to 0.74), Sp =0.93 (0.89 to 0.96).

So the monoclonal EK-CAL performed slightly better than the PhiCAL

Table 28. Comparison of FC tests

Study ID	Number of samples compared	Diseases being diagnosed	Test 1	Test 2	Measurements comparing tests
Burri 2012[86]	361	Organic vs non-organic bowel disease	Phical ELISA >22.4 µg/g Se= 0.66 (0.57 to 0.74) Sp=0.93 (0.89 to 0.96) AUC=0.842	EK-CAL, Buhlman ELISA) >51 µg/g Se=0.78 (0.71 to 0.85) Sp=0.95 (0.92 to 0.97 AUC=0.918	Correlation rho =0.702 Mean (SD) of difference in measurements =30.9 (198.0) µg/g Difference in AUC p<0.001
Kok 2012[33]	382	Organic (including all adenomas as OBD) vs non-organic bowel disease	Quantum Blue POCT 50 µg/g Se=0.64 (0.54-0.72) Sp=0.53 (0.48 - 0.59) AUC=0.66 (0.60 to 0.72)	EK-CAL, Buhlman ELISA 50 µg/g Se=0.74 (0.64-0.82) Sp =0.47 (0.41-0.53) AUC = 0.65 (0.59 to 0.72)	ICC= 0.88 (0.85– 0.90) kappa = 0.66 (0.59–0.73)]
Kok 2012[33]	382	Organic including advanced > 1 cm adeonomas as OBD) vs non-organic bowel disease	Quantum Blue [POCT] 50 µg/g Se=0.76 (0.64 to 0.85) Sp= 0.54 (0.48 to 0.59) AUC=0.75 (0.67 to 0.72)	EK-CAL, Buhlman ELISA 50 µg/g Se= 0.82 (0.70 to 0.91) Sp= 0.45 (0.39 to 0.51) AUC=0.73 (0.66 to 0.81)	
Otten 2008[65]	114	IBD vs IBS	Prevent ID CalDetect POCT 15 µg/g Se= 0.96 (0.78 to 1.00) Sp=0.95 (0.88 to 0.98)	Phical ELISA 50 µg/g Se=0.96 (0.78 to 1.00) Sp=0.87 (0.78 to 0.93)	Cohen's kappa = 0.69
Damms 2008[53]	140	IBD vs non-IBD	Prevista Rapid POCT Se=0.89 (0.65 yo 0.98) Sp=0.80 (0.69 to 0.89) AUC=0.896	EK-CAL Buhlman, ELISA Se=1.00 (0.82 to 1.00) Sp=0.79 (0.67 to 0.98) AUC=0.955	Correlation of line intensity of rapid with ELISA test value r = 0.862

Hence in these studies in newly-presenting patients, there is little difference in performance between the POC tests and the ELISA tests.

Studies comparing FC tests – in studies not in newly presenting patients

These are less useful for our purposes because they may reflect comparative reliability at much higher FC levels in active IBD, or conversely lower levels in those in remission.

Caveat. Many of the comparative studies are not yet published in full so details are often sparse. Missing details include those relating to sponsorship.

Studies comparing POCT and ELISA tests

Kolho 2012 compares QUANTUM Blue (Buhlmann) with ELISA, presumably EK-CAL, in 132 (134?) stool samples from 56 paediatric patients with CD, median age 13 years, range 1-18.[64] Faecal samples were obtained from ten 10 European paediatric gastroenterology units, from patients taking part in European Growth, Relapse and Outcomes With Therapy (GROWTH) CD study.

Thirty of the faecal samples were obtained at the time of diagnosis, and the others, 8 to 72 weeks after starting treatment. The same stool samples were used for both tests.

Median FC value was significantly higher using the QB (317 mg/g [IQR 81–830; range 0–1862] vs 172 mg/g [IQR 50–840; range <30–1656, respectively; P = 0.001]) compared to the ELISA assay. (Note the high levels, a reminder of the need for caution when extrapolating from the results of studies not in newly presenting patients.)

Table 29. Comparison of POCT and ELISA tests in Kolho 2012

	Median FC levels (µg/g)	**IQR**	**Range**	**Correlation - Spearman r**	**ICC analysis**	**Agreement between tests at 100 µg/g cut-off**	**Agreement between tests at 150 µg/g cut-off**
ELISA	172	50-840	<30 - 1656	0.94 p<0.001	0.97 [95% CI 0.95 to 0.98)	87% - kappa 0.87 (95% CI 0.60–0.84).	87%, kappa 0.87
Quantum Blue	317	81–830	0-1862				

The correlation between tests was better in values <300 µg/g, when dilutions were not required, which is the range most relevant to this review. Correlation between tests was high - Spearman r = 0.94, P < 0.001. There was more scatter at higher levels (from figure 1), but this was seen mainly above 600 µg/g.

Interestingly, the authors comment that there was no difference in relative test performance between samples at first presentation and from follow-up (page 437), but note that all patients had CD.

Wassell 2012 compared Quantum Blue POCT against PhiCal (Calpro AS) ELISA, in 47 samples sent to the laboratory for "routine calprotectin analysis".[89] They tested three extractions of the same stool in three patients and found considerable variation: results varied from —31.3% to +31.5%.

Both manufacturers recommend a <50 µg/g cut-off as upper limit of normal. With that cut-off, four of the 47 patients had results that fell on different sides of the cut-off. Two patients were positive by ELISA but negative by POCT and two were negative by ELISA but positive by POCT.

The authors, from Bristol, concluded that Quantum Blue was suitable for excluding inflammatory bowel disease. They suggested that the POCT could be used in GI clinics, to give immediate results, or in smaller laboratories that do not have sufficient throughput to justify an ELISA system.

Dolci 2012 (published as a letter to the editor**)** compared the Quantum Blue POCT test with the Calprest (Eurospital) ELISA assay in stool specimens from 67 consecutive patients with suspected IBD and found a 92.5% (95% CI 83-98) agreement.[90] POCT testing was done on fresh samples. Samples for the ELISA test were frozen, thawed and tested within 2 weeks of collection.

Table 30. Comparison of POCT and ELISA tests in Dolci 2012

	Positive	**Negative**	**Total**
ELISA (cut-off of 90 µg/g)	20	47	
POCT (cut-off of 200 µg/g)	17	50	67

Note that the cut-off used for the Quantum Blue POCT was much higher than that of the established ELISA method. Five patients showed discrepant results, four being positive only with ELISA (two borderline results, 94 and 98 µg/g stool) and one positive only with POCT.

Coorevits 2012 (abstract only**)** compared Quantum Blue POCT with the Buhlmann ELISA in 128 samples, in patients aged from 16 to 72. [91]

Cut-off values used were;

- Negative for IBD - < 50 µ g/g faeces (as suggested by manufacturer).
- Positive for IBD - > 200 µ g/g faeces

- Intermediate zone 50-200 µg/g faeces, result uncertain

Coorevits et al noted that FC values up to 210 µg/g faeces have been described in IBS patients, and used an ELISA cut-off of >200 as indicative of IBD. They had 50 patients with results above this level and 83 below.

They found good correlation ($R^2 = 0.89$) between the tests. After applying different cut-offs to the POCT and assessing the numbers of discordant results between POCT and ELISA, they concluded that 30 µg/g for ruling out inflammation, and 110 µ g/g for confirming it, appeared to be the most suitable cut-offs for the POCT. This left a grey zone of 30 to 110. This gave 89.4 % (127/142) agreement with the ELISA and 10.6 % (15/142) mismatches.

Studies comparing different POCT tests.

Hessells 2012 compared two rapid tests, Quantum Blue and Prevent ID CalDetect, using the laboratory quantitative time-resolved fluorimetric immunoassay (TRFIA) as the gold standard, using a cut-off of 50 µg/g.[92] The Prevent ID is a rapid semi-quantitative test, with lines: negative, < 15 µg/g. has two lines; positive, >60 µg/g, has four, and indeterminate, 15-60, has three.

The TRFIA test is reported to have some advantages over ELISA (better precision, wider range, greater sensitivity), but need not be considered further here – its role is simply as gold standard comparator for the two rapid tests.

The patient group was a mixture of new referrals with suspected IBD (n=40), and suspected relapses (n = 45) referred to a Dutch gastroenterology unit. Performance was assessed at four cut-off levels for Quantum Blue (30, 40, 50 and 60 µ g/g) and two cut-off levels for CalDetect (15 µ g/g and 60 µ g/g) [the lowest and highest detection levels}. The same samples were used for rapid and TRFIA testing. A TRFIA level of 50 µ g/g was used as the golden standard test performance.

Table 31. Comparison of tests in Hessells 2012

	Cut-off level -Quantum Blue µg/g	Cut-off level - TRFIA µg/g	Correct classification	Se	Sp	PPV	NPV
Quantum Blue	30	50	0.77	0.96	0.69	0.55	0.98
	40	50	0.86	0.92	0.84	0.69	0.96
	50	50	0.88	0.88	0.84	0.68	0.94
	60	50	0.85	0.79	0.87	0.7	0.91
	Cut-off level - ELISA CalDetect						
CalDetect	15	50	0.65	0.96	0.53	0.44	0.97
	60	50	0.78	0.88	0.74	0.57	0.94

Optimal cut-off levels were 40 µg/g for the Quantum Blue test (negative predictive value 0.96, sensitivity 0.92, specificity 0.69) and 15 µg/g for the Caldetect test (negative predictive value 0.97, sensitivity 0.96, specificity 0.44).

The correlation between the rapid tests and TRFIA was good for both tests (kappa test; $p<0.0001$), but significantly better for Quantum Blue (kappa 0.77; 95 % CI 0.64 – 0.90) than for CalDetect (kappa 0.46; 95 % CI 0.32 – 0.60).

The authors concluded both tests performed well, but that the Quantum Blue test was superior (at cut-off 40 µg/g) to the Prevent ID CalDetect in reducing the number of colonoscopies. Because of its high NPV, the number of colonoscopies might be reduced by 62%. The Quantum Blue can be used with a POC reader giving a quantitative result.

Studies comparing POCT with ELISA

Sydora 2012 also compared the Quantum Blue with a standard calprotectin ELISA method (from Alpco Immunoassays Salem - probably Buhlmann).[93] The participants included patients with UC, CD or IBS, and volunteers with no known intestinal problems.

The IBD patients group had significantly higher calprotectin levels than IBS patients and healthy controls ($p=0.01$). There was no difference in calprotectin concentrations between IBS patients and controls. Some IBD who had had recent surgery had calprotectin levels similar to controls and IBS patients.

Results were available in 8 hours from the ELISA method but in 30 minutes from Quantum Blue. However the ELISA had a much wider range. Quantum Blue has a minimum measurement value of 30 μg/g and a maximum of 300 μg/g. This would not be a problem for the NICE decision group of patients, where the focus is in distinguishing between normal and raised levels. So as soon as the level is abnormal, referral is triggered and the height does not matter at this stage.

Sydora et al concluded that;

- with Quantum Blue, a cut-off at 150 μg/g distinguishes healthy control subjects and IBS patients from those with active IBD with a specificity of 100%. (After excluding IBD patients who had had recent surgery)
- ELISA testing gives the same specificity at a cut-off of 230 μg/g
- The desk-top Quantum Blue is as accurate as the ELISA in distinguishing between inflammatory and non-inflammatory intestinal disorders but can do so in 30 minutes compared to 8 hours for the ELISA.

Vestergaard 2008 compared the semi-quantitative PreventID Caldetect with the PhiCal ELISA (Calpro,Oslo) in 95 samples from 82 patients and 13 healthy volunteers with no history or symptoms of bowel disease.[94] The patients had IBD (27 CD, 15 UC, 3 indeterminate colitis); chronic diarrhoea (24); abdominal pain (6) or other reasons. The age range of the patients was 2–86 years. Their results are shown in table x.

	Sensitivity % ELISA cut-off 50	**Specificity % ELISA cut-off 50**	**PPV %**	**NPV %**
Rapid test, cut-off 60 μg/g	66 (52-79)	100 (53-80)	100 (90-100)	72 (60 - 83)
Rapid test, cut-off 15 μg/g	96 (87-100)	70 (55-83)	79 (67-88)	94 (81-99)

Correlation was good but 18 patients had a positive calprotectin ELISA test but were negative with the rapid test. The authors used the recommended ELISA cut-off of 50 mg calprotectin/kg stool.

Vestergaard et al regarded a calprotectin concentration by the rapid test of <15 μg/g as reliable for excluding IBD. With calprotectin concentrations >15 μg/g, they recommend checking the POCT result by quantitative measurement. So the Prevent ID could be a useful screening test to rule out inflammation.

Shastri 2009 compared the Immundiagnostik ELISA with the PreventID CalDetect, with cut-off 15 ng/mL for both tests, in 823 patients. [95] The ELISA had slightly better sensitivity and specificity than the POCT (e.g. Se for CD 96% versus 93% , Sp 89% versus 83%) but these were not significantly

different. For NPV the different was greater (88% versus 79% for CD; 83% vs 76% for UC) but again, CIs overlapped.

Shastri et al report that the POCT can be done in 5 minutes, which is less than most reports.

Labaere et al compared eight different assays for calprotectin: four ELISAs, three POC (Quantum Blue, Eurospital Calfast, Biotest Certest) and the automated immunoassay from Phadia (details available only from a recent meeting abstract [96]) and poster. They compared the tests for both distinguishing IBD from non-IBD, and for monitoring IBD, and also compared results with endoscopic and histological findings. They reported that sensitivity (82 to 83%) and specificity (84 to 89%) were similar amongst the assays. For distinguishing IBD from non-IBD, they concluded that the best tests were Quantum Blue, Phadia and Calprolab, with ratios of median IBD to non-IBD of 14, 12 and 10, They conclude that;
"All calprotectin assays showed acceptable and comparable clinical performance for diagnosis of IBD".

They conclude that the quantitative POCTs could replace the ELISA tests. They had reservations about the comparative merits of the tests for monitoring disease activity, but that is not relevant to this review.

Summary
The overall message from these studies is that the POC tests are about as good as the ELISA tests.

Studies comparing ELISA tests
Whitehead et al[97] compared three ELISA tests, Immundiagnostik, Buhlmann and Eurospital. All assays performed satisfactorily, but the Buhlmann gave higher results. They suggests that each laboratory determine its own reference range.

Loitsch 2010 [meeting abstract]
The tests compared were Immundiagnostics (Calp-ID) and Buehlmann EK-CAL (Calp-Bu).[98]

The patients were a mixture of those with active IBD, those with JBD in remission, and those with IBS. There were 108 patients with IBD (77 active and 31 in remission), and 96 with IBS. Loitsch and collages used the manufacturer's cut-off values. The sensitivities, specificities and accuracy of the and the Buhlmann EK-CAL were as shown. Note that the Sp appears to be the same in all three groups but other parameters vary.

Table 32. Comparison of tests in Loitsch 2012

	Specificity Calp-Bu	Specificity Calp-ID	Sensitivity Calp-Bu	Sensitivity Calp-ID	Accuracy Calp-Bu	Accuracy Calp-ID
IBS (n=96) vs active IBD (n=77)	63.40%	79.20%	97.40%	93.50%	78.60%	82.10%
IBS (n=96) vs active colitis(n=77)	63.40%	79.20%	100.00%	97.80%	75.30%	85.20%
IBS (n=96) vs active CD (n=41)	63.40%	79.20%	95.40%	90.20%	72.90%	82.50%

The specificity of Calp-ID is higher and sensitivity is lower, but the overall accuracy is higher. Note the much lower specificity for both tests. The authors concluded that both tests provide a reliable and non-invasive way of differentiating IBD from IBS, but that the Calp-ID was the more accurate test.

Tomkins 2012 [meeting abstract]

Tomkins et al from Coventry compared two ELISAs, Immundiagnostik PhiCal (version 1) and the Buhlmann EK-CAL in 62 patients, of which 38 had IBD or other organic pathology (age range 15-49, mean 36), and 24 had IBS (age range 20-48, mean 36).[99] All participants had a colonoscopy with biopsy.

Table 33. Comparison of tests in Tomkins 2012

Test	Cut-off value	Se (95% CI)	Sp (95% CI)	PPV (95% CI)	NPV (95% CI)
Buhlmann EK-CAL	50 µg/g	86% (42 to 99)	60% (33 to 83)	50% (22 to 78)	90% (54 to 99)
PhiCal1	50 µg/g	78% (40 to 96)	92% (60 to 100)	88% (47 to 99)	86% (56to 97)

Hence using 50 µg/g as the cut-off, PhiCal 1 performed slightly better than EK-Cal but NPVs were similar.

Results from different FC methods are not directly comparable, despite widespread adoption of single cut-offs.

Section 2.11 has been updated. Please refer to the Diagnostics Assessment Report Addendum

2.11 GP assessment and referral: implications for modelling.

Adults

As noted previously, we lack published data on the use of calprotectin testing in primary care. However we have the unpublished results from the NTAC pilots, and these provide data on referral patterns by GPs in the UK (assuming that those in the North-East are representative).

The Durham Dale pilot provides data on GP referrals with no calprotectin testing, and the effect that testing would have. The data allow us to explore what might happen if calprotectin testing is made available.

The test used was the POCT Prevent ID, which divides people into 3 groups;

- Negative < 15µg/g
- Positive > 60 µg/g
- Intermediate >15 but < 60

GPs made referral decisions based on clinical assessment without knowledge of the calprotectin results. They referred those that they thought might have IBD, and managed those that they thought had IBS in primary care.

A final consultant diagnosis was made, based on calprotectin test results and clinical data, including endoscopy. The clinical data came from GP and OP data, where patients were referred, or just from GP data, when patients were not referred. Note that those diagnosed as IBS (and not referred) did not have colonoscopy so it is not possible to completely exclude false negatives. These would have IBD but appear clinically to be IBS and have negative calprotectin results. Such false negatives are unlikely given the high sensitivity (100% - see figure 3) of calprotectin in this POCT at the 15 µg/g cut-off, but not impossible. (The Durham Dale pilot could not be used in our main assessment because of the lack of a definitive reference test.)

For assessing the sensitivity and specificity of GP assessment, there are two options using the Durham Dale pilot data.

1. Use calprotectin as reference test.
2. Use final consultant diagnosis.

If we compare GP diagnosis with calprotectin levels, and assume that a positive calprotectin test implied possible IBD and an indication to refer, then we have a 2x2 table as follows

Section 2.11 has been updated. Please refer to the Diagnostics Assessment Report Addendum

Table 34. GP diagnosis compared to calprotectin level.

	FC +ve	FC –ve	Total
GP IBD	28	4	32
GP IBS	6 (4 high, 2 indet.)	79	85
	34	83	117

So Se 28/34 = 82% and Sp 79/83 = 95% where "positive" = a positive FC test. If we exclude the two indeterminates (who would be re-tested, rather than referred), sensitivity is 88%. Of the 83 diagnosed as IBS, only 4 had high calprotectin, a 5% error rate giving NPV of 95%. Note that the four are not false negative in the sense of being missed IBD, but in the sense of being "false non-referrals". Not all would have IBD.So without FC testing, GPs would not refer four of 32 patients with high calprotectin

This means that the consultant diagnosis is more useful for our purposes, and the next table compares the GP diagnosis (without knowledge of calprotectin result) and the consultant diagnosis (with knowledge of calprotectin result and of endoscopy where performed. Note that far more patients (33) had endoscopy than were found to have IBD.)

Table 35. GP diagnosis compared to final consultant diagnosis

	Consultant IBD	Consultant IBS	Total
GP IBD	7	22	29
GP IBS	0	82	82
	7	104	111

Numbers are slightly less than in the previous table because some patients do not appear to have been followed up. No data are given in the YHEC report on the presumed diagnosis or calprotectin results in five missing cases. The sixth was found to have cancer.

These results show that the GPs referred all those diagnosed as IBD, giving a "whole pathway" sensitivity of 100% (if we assume there were no false negative IBDs as discussed above.) "Whole pathway" combines GP assessment, calprotectin testing and consultant opinion based on clinical data that included endoscopy (mainly colonoscopy but some flexible sigmoidoscopies).

However this is achieved at a specificity of 79% for GP assessment without calprotectin testing. Without calprotectin testing, GPs refer a group of whom around 25% have IBD (7 of 29) and 75% have IBS. This matches results from routine care that over 60% of colonoscopies in young people are normal.

This implies that if GPs had access to calprotectin testing, they might be able to reduce referrals by a considerable amount – about three-quarters. The Durham Dale data suggest that GPs refer about a quarter of patients presenting to them with gastrointestinal symptoms that could be due to IBS or IBD.

The number of patients is quite small, but that proportion is similar to the figure of 29% reported in the BSG guideline on IBS, which increases confidence in the pilot data.[100]
The prevalence of IBD in the whole population was 6.3 % (SE 2.3%), but amongst those referred, it was almost 25% (6.3 of 25%.

In the pilot, a GP decision to refer set a patient on a pathway that could lead to colonoscopy and possible other invasive investigations. This decision would not be taken lightly. However, if faecal calprotectin testing is introduced, we might expect that GPs would consider testing in a wider patient group then they would consider for referral. They refer only about 25% of those that present to them with these symptoms. We can create a scenario analysis, assuming that if calprotectin testing becomes available, GPs will test twice as many as they would have referred in the absence of faecal calprotectin testing.

We also note from the Durham pilot that if GPs thought that a patient had IBS, they were right at least 95% of the time because only 5% of those they thought had IBS had high calprotectin and needed referred. (NPV 95%).These "false non-referrals" could theoretically include some with IBD.
In our scenario analysis, we assume that all patients with IBD will be in the larger group (50% of all patients with symptoms, so 222 patients) that will have calprotectin testing. If we assume that 50% of patients with symptoms will be tested, we get figures as shown in table 36. All of the 6.25% of patients with IBD are tested, and assuming the POCT sensitivity of 100%, no patients with IBD would be missed.

If we used an ELISA test, with a sensitivity of 93% (from meta-analysis, we would miss 0.44%, or 0.49 patients in the numbers in this group.

The extra group are those regarded by the GP as less likely to have IBD than the 25% (because the GP didn't refer them), and GP is really doing the test to confirm IBS. The false positive rate amongst the additional 25% tested, will therefore be much less than in the 25% referred. One option is to assume that there will be no new false postives
So figures change to;

Table 36. Expected numbers if 50% of presenting patients are tested with FC.

	IBD	IBS	
GP + FC IBD	7	22	
GP + FC IBS	0	193	
	7	215	222

The prevalence of IBD in the tested group is half that in the referred group – about 12.5%. Since all those with IBD are tested, the false negatives if we assume sensitivity of calprotectin testing to be

100%. Specificity is 90%. If we assumed that there would be more false positives, specificity would be 80% if we double the false positives to 44 and 85% if we increased them to 33.

If the calprotectin test was the average ELISA with Se 93% and Sp 94%, the figures in the above table would change to;

Table 36b

	IBD	IBS	
GP + FC IBD	6.51	13.19	
GP + FC IBS	0.49	201.51	
	7	215	222

Only 9% would be referred due to the greater Sp of ELISA, but 0.49 patients would be missed.
If we assume that only patients with raised calprotectin are referred, and that calprotectin is 100% sensitive for detecting newly presenting (and hence active) IBD, then with calprotectin testing, GPs will refer about 13% compared to the 25% referred when they have no calprotectin testing available – a drop of around half%. However, not all the calprotectin false positives would be referred if GPs, aware of the imperfect specificity of the test, used clinical judgement and a repeat test with the more specific (94%) ELISA test before referral. That would reduce number referred to about 20 (approx. 7TPs and 13 FPs) or 9% - a drop of over 60%

So for modelling purposes, using the Prevent ID test , we can use a prevalence of IBD of 6.3% , and in the absence of faecal calprotectin testing, a sensitivity of GP referral of IBD of 100%, and 79% specificity.

Using the North European data from Shivananda et al[25], we would expect in this adult group, a ratio of UC to CD of 3:2. (Incidence of UC 12.9 in 15-44 age group, based on 539 cases; of CD 8.7, based on 365 cases.).

Note that there are some weaknesses in the above arguments;

1. The 50% is a rather arbitrary assumption. We have reasonably assumed that more patients with symptoms would have calprotectin testing than were referred when testing was not available, but we cannot say if 50% is correct. Given that GPs are good at diagnosing IBS, we would not expect 100% to be tested.
2. Our base case assumption is that doubling the number tested would not increase the number of false positives. Since the extra 25% tested would have less severe symptoms than the first 25% (referred), it seems reasonable to rule out a doubling of false positives. However assuming no increase may be too optimistic

Section 2.11 has been updated. Please refer to the Diagnostics Assessment Report Addendum

3. The 100% sensitivity for the POCT test is based on only one study with not very large numbers, and needs to be replicated in a larger study. The mean ELISA Se was 93%. However, GPs would not simply rely on the test results alone, knowing that sensitivity was not perfect, and some of the false negatives on ELISA testing might be referred on clinical nous.

Children

Modelling requires different assumptions in children. Based on the recent UK study by Henderson et al, 48% of referred cases (91/190) had IBD.[26] The ratio of CD to UC is much higher – 2.3:1. The potential reduction in colonoscopies is therefore greater.

3. Economics

In the following all costs have been converted to 2011 prices using the PSSRU HCSC price index.[101] Any foreign currency amounts have been converted to sterling at the contemporaneous April 05 exchange rate, with these amounts then being converted to 2011 prices using the PSSRU HCSC price index. Where the base year is not given within the paper, it has been assumed to be the year of publication. The original amounts are given in square brackets.

A review of the cost effectiveness literature for faecal calprotectin testing is presented. This is followed by a review of studies of quality of life that may suitable for inclusion in a cost utility analysis of faecal calprotectin testing, health related quality of life for three conditions having to be considered: IBS, Crohn's disease and ulcerative colitis. Given the centrality of colonoscopy to the question in hand, a brief review of the adverse events associated with colonoscopy is then presented. A relatively simple cost consequence model of faecal calprotectin testing is then presented, augmented by some considerations around the loss of utility among false negatives during their period of incorrect treatment. This is followed by a full cost utility model, much of the structure of this being drawn from the modelling for *CG61: Diagnosis and management of irritable bowel syndrome in primary care*;[35] the modelling for *CG152 : Crohn's disease: Management in adults, children and young people*;[102] and the modelling for the current draft of the ulcerative colitis guideline: *Ulcerative colitis: management in adults, children and young people*.[103]

FC tests economic literature

Hornung and Anwar analysed the results of the 40 patients who had faecal calprotectin testing between January 2009 and April 2010.[104] This appears to be all the patients tested with faecal calprotectin within the North Tees and Hartlepool NHS Foundation Trust. No detail is given of which calprotectin method was used. Patients were split into those with IBS like symptoms of unknown cause in whom IBD needed to be ruled out (n=22) and those with known IBD (n=18). 9% (two people) of the first group and 61% of the second group had a high (level not stated) calprotectin result. But Hurnung et al note that in the first group faecal calprotectin testing did not result in a change in treatment for any patient, compared to 12 of the 18 IBD patients having their treatment changed as a result of the faecal calprotectin result. As a consequence, it appears that of the 8 colonoscopies avoided, none were in the group of patients with IBS like symptoms of unknown cause in whom IBD needed to be ruled out. However it is reported that 13 of the 17 colonoscopies in the newly-presenting group were normal, though it is not clear if the four with abnormal findings had IBD, nor if they included the 2 newly-presenting patients with high calprotectin.

Mindemark and Larrson undertook a cost minimisation analysis comparing the diagnostic pathway of faecal calprotectin testing followed by colonoscopy with direct referral to colonoscopy, the aim being to rule out IBD.[105] For faecal calprotectin two cut-offs were used: 50µg/g and 100µg/g. The study data was drawn from a retrospective analysis of 3,639 Swedish patients. Test costs were £24.57 [€29] for faecal calprotectin, £576.20 [€680] for colonoscopy in adults and £1,152.40 [€1,360] for colonoscopy in paediatrics. One third of patients were paediatric, with a further 13% being aged over 65. In the paediatric group 54% had a faecal calprotectin of < 50µg/g, while 71% were < 100µg/g. In those aged 18-65 52% had a faecal calprotectin of < 50µg/g, while 68% were < 100µg/g, with the respective percentages for those over 65 being 30% and 51%. If a threshold of 50 µg/g was used to exclude organic disease, around 50% of colonoscopies could be avoided. If a cut-off of 100 µg /g was used, 67% could be avoided.

The direct costs of the diagnostic strategies were £2,791,680 [€3,294,600] for direct referral to colonoscopy, £1,461,369 [€1,724,611] for faecal calprotectin with a 50µg/g cut-off and £985,409 [€1,162,931] for faecal calprotectin with a 100µg/g cut-off. The study does not consider false negatives and the costs of them subsequently re-presenting. Nor does it specify what assumptions were made about sensitivity and specificity – it appears that perfect distinction between inflammatory and non-inflammatory is assumed.

One useful point made is that a reduction in colonoscopy for distinguishing between inflammatory and functional conditions would help resolve waiting list pressures for colonoscopy for other reasons. This is relevant to the UK following the roll-out of colorectal cancer screening. Calprotectin testing may not save money since the capacity released might be used for other purposes, but it would reduce or avoid the need to expand services to cope with, for example, follow-up after colorectal screening.

Goldfarb et al compared wireless capsule endoscopy with colonoscopy coupled with a small bowel follow through with a barium swallow for the diagnosis of Crohn's disease.[106] They note that while 75% of Crohn's disease patients have small bowel involvement, more than one third have disease limited to the small bowel. The parameters populating the decision tree model are not entirely clear, but it also included a perforation rate for colonoscopies of 0.03% and retention of the wireless capsule in 0.75% of cases. Costs were based upon Medicare reimbursement rates, with the conclusion that wireless capsule endoscopy had a diagnostic yield of 70% compared to 54% for colonoscopy with SBFT, while also saving an average £197 [$291].

The YHEC faecal calprotectin testing report for the Centre for Evidence Based Purchasing provides the most comprehensive review to date of the economics of faecal calprotectin testing compared to testing with ESR and CRP.[44] It considers the primary care patient population, presenting with

symptoms suggestive of IBS but no "red flag" symptoms. Those testing positive are assumed to be referred to secondary care for colonoscopy. Those testing negative are treated as IBS patients, initially with dietary and lifestyle advice. Among those testing negative, for those with IBS 50% do not respond to dietary and lifestyle advice while for those with IBD 100% of do not respond to dietary and lifestyle advice. Non-responders seek further advice and medication from their GP after two months. Among non-responders, for those with IBS 5% do not respond to further medication while for those with IBD 100% of do not respond to further medication. These are then referred on to secondary care for further investigation. For the base case, all investigations are assumed to be colonoscopy with no sigmoidoscopy though both are assumed to have 100% sensitivity and 100% specificity.

Table 37. YHEC report base case parameter values

Presenting population					
Prevalence of IBD in presenting population					10%
IBS patients uncontrolled on dietary advice alone					50%
Of whom uncontrolled on medication and requiring further investigation					5%
Test characteristics					
	ESR/CRP	FC	FC ELISA	FC POCT	Colonoscopy
sensitivity	35%	90%	96%	61%	100%
specificity	73%	80%	87%	98%	100%
cost	£4.64	£25.00	£25.00	£27.68	£544

For the base case comparing ESR/CRP against FC, FC was found to be dominant due to FC correctly diagnosing more IBS patients and more IBD patients at lower cost. Note that within the model all false negatives eventually re-present and as a consequence all IBD patients are eventually correctly diagnosed.

A second comparison comparing FC ELISA with FC POCT found that FC ELISA was more expensive overall. While FC ELISA diagnosed more IBD patients correctly, due to its poorer specificity it also resulted in more IBS patients being incorrectly sent for colonoscopies. This was the source of the additional costs under FC ELISA. This underlines the importance of the specificity of the tests, particularly given the relatively low prevalence of IBD in the presenting patient population.

Results were sensitive to the prevalence of IBD in the presenting population, sensitivities and specificities and the costs of the tests.

Mascialino et al[a] augment the YHEC model with a third branch in the decision tree model for faecal calprotectin for indeterminate results of between 50µg/g and 250µg/g.[107] Those with faecal calprotectin > 250µg/g follow the YHEC positive result branch, those with faecal calprotectin <

[a] Sponsored byThermo Fischer Scientific

50µg/g follow the YHEC negative result branch, while those with an indeterminate result receive a second test. Unfortunately, possibly due to only being a poster presentation, quite how the new indeterminate branch is populated is unclear. The tables of the poster still only report sensitivities and specificities. The overall conclusions mirror those of the YHEC report, only with more correct diagnoses and larger cost savings. But it is unclear how these have been arrived at due to the lack of detail about how the third branch of the model has been populated.

In another conference abstract, Mascialino et al report that in modelling, calprotectin dominates CRP and ESR being more accurate and less costly with an estimate of £100 lower cost per patient in the UK.[45]

Dubinsky et al modelled the cost effectiveness of three main alternative diagnostic strategies in the US context, using serological markers: anti-Saccharamyces cerevisiae antibodies (ASCA) for Crohn's disease and perinuclear antineutrophil cytoplasmic antibodies (pANCA) for ulcerative colitis.[108] They developed a decision tree model comparing immediate referral for colonoscopy, with the possibility of barium upper GI investigation and a small bowel follow through, with two diagnostic testing strategies: a primary ASCA and PANCA assay with subsequent referral to colonoscopy and a sensitive primary assay followed by a more specific second confirmatory assay with subsequent referral to colonoscopy. Costs were taken from the Medicare fees schedule. Those with negative results could return for further testing after 2-3 months, with 50% of true negatives with IBS representing and presumably 100% of false negatives with IBD representing. Unusually, based upon expert opinion, Dubinsky et al do not assume 100% sensitivity and 100% specificity for colonoscopy.

Table 38. Dubinsky model inputs

Presenting population			
Prevalence of IBD in presenting population			20%
IBS patients with persistent symptoms			50%
Test characteristics			
	Primary Assay	Second Assay	Colonoscopy
sensitivity	80%	65%	95%
specificity	50%	65%	95%
cost	£48 [$54]	£48 [$54]	£1,730 [$1,880]

Cost effectiveness over a one year period was measured as the cost per correct diagnosis, this encompassing both correct diagnoses of IBD and correct diagnoses of IBS. Given this definition of effectiveness, the sequential testing strategy resulted in more correct diagnoses that both direct referral and a single primary test before referral: 97.90% accuracy compared to 95.95% and 96.95% respectively. The sequential testing was also cheaper than both direct referral and a single primary test before referral: £1,511 [$1,641] compared to £2,015 [$2,189] and £1,740 [$1,890] respectively. As a

consequence, the sequential testing was found to dominate both direct referral and a single primary test before referral. The results for sequential testing are of interest, though their relevance is limited to a degree by effectiveness being measured in terms of correct diagnoses and so not distinguishing between correct diagnoses of IBS and correct diagnoses of IBD. The relevance of the results is limited by ASCA and pANCA not being regarded as comparators in the NICE scope, but it does illustrate the possible benefits of sequencing tests.

Quality of Life

HRQoL studies have been summarised if they provide direct estimates of utilities, or provide supporting data on either the differences in quality of life between any of the conditions under consideration or data on the quality of life related to symptom severity. Patient characteristics in terms of age, sex, disease severity and disease duration are not presented in the text but to the extent they are available are presented in the summary tables.

IBS: Quality of life studies

Akehurst et al [b] undertook a survey of IBS patients in the UK primary care setting. A sample of 161 patients with IBS was selected from GP lists based upon the Rome I criteria, with an additional 213 control patients being selected.[5] Controls were matched for age, sex and social characteristics by the patient's GP. The SF-36, EQ-5D and IBS-QOL questionnaire were administered at baseline and subsequently at 3 months. Patients with IBS reported average baseline SF-36 values that were statistically significantly worse than those reported by the control group for every dimension of the SF-36. Similarly, for the EQ-5D the mean baseline score reported by those with IBS of 67.5 was statistically significantly lower than that of the control group. Unfortunately, it is not clear what EQ-5D rating algorithm was used for this with it being described as the "*EQ-5D derived score*", and there is no reference to the UK social tariff. The mean baseline EQ-5D rating scale, presumably the EQ-5D VAS, reported by those with IBS of 64.2 was also statistically significantly worse than the 80.3 of the control group. Parallel statistically significant differences were also reported at 3 months, though the mean quality of life values between baseline and three months were not statistically different.

In what was apparently a follow up study to Akehurst et al, Ricci et al (2003)[c] compared the HRQoL of 305 IBS patients selected from GP lists with 330 controls.[109] Unfortunately, Ricci et al is only available as an abstract, but notes a statistically significant relationship between the severity of IBS and patients' reported VAS scores. No further detail on this is provided.

[b] Sponsored by Novartis
[c] Sponsored by Novartis

Bernklev also reports SF-36 scores among IBS patients and compares these with French population norms, finding IBS to significantly adversely affect scores, but no overall quality of life values are reported.[110]

Within a broader paper comparing the performance of the EQ-5D and SF-36 across seven patient groups, Brazier et al apply the EQ-5D and SF-36 to 161 UK IBS patients recruited from primary care.[111] All patients were observed twice. The EQ-5D was evaluated using the UK social tariff, while the SF-35 was valued using the SF-6D algorithm. Based upon 314 responses the mean EQ-5D index was 0.662, compared to 0.666 for the SF-6D. It appears probable given the similarity of baseline characteristics and results with those of Akehurst et al (and that Brazier was a named author of the Akehurst et al paper, that the IBS patient group and responses were the same for both papers.

Bracco et al[d] in an economic evaluation of tegaserod compared to placebo for IBS evaluated the EQ-5D responses using the UK social tariff of IBS patients: 247 receiving tegaserod and 238 receiving placebo.[112] The adjusted average baseline utility was 0.726. At week 4 these had improved to 0.795 in the tegaserod group and 0.759 in the placebo group, but by week 12 had fallen back slightly to 0.792 and 0.747 respectively.

DiBoneventura et al[e] compared SF-6D utilities among 109, 83 and 204 patients with IBS-C in the UK, France and Italy respectively with matched controls in the UK, France and Italy.[113] Respondents were recruited through the National Health and Wellness Survey, a self-administered internet based survey. UK patients with IBS-C reported a mean utility of 0.65 compared to 0.71 for their matched controls, the corresponding figures being 0.63 and 0.71 for the French sample and 0.66 and 0.70 for the Italian sample.

Pare et al[f] (2006) reported the UK social tariff EQ-5D index among 1,555 Canadian primary care patients.[114] The patient group recruited had mainly IBS-C or IBS-A, apparently due to a desire for the results to be relevant to patients eligible for tegaserod. The mean EQ-5D index was 0.64.

Puhan et al applied time trade off, standard gamble and the SF-36 to 96 Canadian patients with IBS. Patients were identified through either medical records of the gastroenterology clinic at McMaster Health Sciences Centre or through 10 local gastroenterologists.[115] The SF-36 was valued using the SF-6D transformation. This resulted in three mean estimates for health related quality of life: 0.84 for standard gamble, 0.76 for time trade off and 0.85 for SF-6D.

[d] Sponsored by Novartis
[e] Sponsored by Novartis
[f] Sponsored by Novartis

Spiegel et al[g] administered the EQ-5D among 257 American IBS patients, with these split into three groups of those with constipation IBS-C, those with diarrhoea IBS-D and those with mixed, IBS-M.[3] Note that the severity of disease within the sample was mixed, with 16% having mild disease, 32% having moderate disease and 55% having severe disease. Patients were followed up at 3 months. It is unclear what algorithm was used to construct the EQ-5D utilities. There was no statistically significant difference in the mean utilities of 0.76 for those with IBS-C, 0.76 for IBS-D and 0.73 for IBS-M. There was a statistically significant difference between the mean utilities of 0.70 for those with severe disease compared to 0.80 for those with non-severe disease, and between the mean utilities of 0.78 for those experiencing considerable relief of symptoms at 3 months compared to 0.73 without considerable relief of symptoms at 3 months.

Wang et al administered the EQ-5D among 198 people with IBS and 251 people without IBS.[2] These were recruited from those attending the National Foundation for Digestive Diseases Symposium, a free public education symposium held at the Raffles hotel in Singapore. The EQ-5D was evaluated using the UK social tariff. The mean utility among those with IBS was 0.739, which was statistically significantly lower than the 0.849 of those without IBS.

IBD: Quality of life studies

Konig et al recruited 121 outpatients and 31 inpatients with IBD from German hospitals, with 123 having Crohn's disease and 29 having ulcerative colitis.[116] Both groups had a mean of around 2 active phases in the past year, with 60% of Crohn's patients in remission compared to 70% of ulcerative colitis patients. 79% of outpatients were in remission compared to only 7% of inpatients. The German version of the EQ-5D was administered. The EQ-5D utility index was calculated using the German population mapping function, which asked respondents to rate EQ-5D health states using the EQ-5D VAS. Konig et al found that 30% of outpatients and 19% of inpatients classed themselves as having health state 11111: i.e. having no problems in all EQ-5D dimensions. 64% of outpatients and 45% of inpatients classed themselves as having no problems in four dimensions, with only one dimension being classed as having some problems. A histogram of the EQ-5D VAS shows a steady increase in the proportion of patients classifying themselves over the range 0 to 80 and then tailing off again, but the EQ-5D index shows very few respondents classifying themselves as having a utility of less than 50. But note that the German EQ-5D index algorithm apparently tends to value health states more highly than the UK social tariff, with this applying with particular force to worse health states. The

[g] Sponsored by Takeda

mean EQ-5D index was 0.875 for those in remission compared to 0.627 for those with active disease, while for outpatients it was 0.803 compared to 0.619 for inpatients.

Leidl et al administered the EQ-5D among 270 patients with Crohn's disease and 232 patients with ulcerative colitis.[117] Patients were recruited from the German Patients' association for inflammatory diseases, and were split into slight (CDAI 0-3), moderate (CDAI 4-7) and severe (CDAI >7) subgroups. Leidl et al also applied both the German and the UK social tariff in their valuations. The mean value for both tariffs were broadly similar, with the exception of Crohn's disease patiens with severe disease who were assigned a quality of life of around 0.50 using the German tariff, but 0.33 using the UK social tariff. The mean values applying the UK social tariff, taken from the graph, for mild, moderate and severe disease were 0.87, 0.67 and 0.33 among Crohn's disease patients, and 0.91, 0.73 and 0.67 for ulcerative colitis patients.

Stark et al contacted a random sample of 724 patients with Crohn's disease and 723 patients with ulcerative colitis from the German IBD association (DCCV), the largest voluntary support organisation of IBD patients in Germany.[118] 37% agreed to participate and 36% completed the EQ-5D at baseline: 270 patients with Crohn's disease and 253 patients with ulcerative colitis. Those with inactive, slight, moderate and severe disease were 57.1%, 33.2%, 9.3% and 0.4% among Crohn's disease patients with 57.1% being in remission, compared to 62.1%, 26.3%, 9.4% and 2.2% for ulcerative colitis patients with 62.1% being in remission. At baseline the mean utility among Crohn's disease patients using the UK social tariff was 0.77 overall, with 0.89 for those in remission and 0.61 for those with active disease. Using the German tariff it was 0.86 overall, with 0.95 for those in remission and 0.75 for those with active disease. At baseline the mean utility among ulcerative colitis patients using the UK social tariff was 0.84 overall, with 0.91 for those in remission and 0.71 for those with active disease. Using the German tariff it was 0.92 overall, with 0.96 for those in remission and 0.84 for those with active disease. The study also re-administered the EQ-5D at 4 weeks, but the resulting data is only used to assess construct validity and is of limited interest for current purposes.

Turunen et al mailed 550 Finnish "paediatric" IBD patients and 1,650 age and sex matched controls from the same municipality a bespoke questionnaire.[119] 67% of the patient group and 37% of the control group responded. The IBD patients had been previously identified for another study, through chart review of 2 major Finnish hospitals. Unfortunately, this resulted in a mean age among responders of 21 years. The questionnaire posed 4 generic questions on physical, emotional, social and overall quality of life with these being rated on a visual analogue scale of range 1 to 7. The main result of interest is that there were no major differences in mean responses between those with Crohn's disease and those with ulcerative colitis.

Casellas et al administered the IBDQ and the EQ-5D among 1,156 Spanish IBD patients, 628 with Crohn's disease and 528 with ulcerative colitis.[120] These were composed of both inpatients and outpatients: 141 and 487 respectively for Crohn's disease, and 108 and 420 for ulcerative colitis. Among Crohn's disease patients 268 were in relapse while 360 were in remission, while among ulcerative colitis patients 212 were in relapse and 316 were in remission. It appears that the valuation of the EQ-5D used the Spanish valuation set as reported in Badia et al.[121] Within a multivariate regression analysis of the IBDQ Cassellas et al found that the underlying condition was not statistically significant, with a t-statistic of only -0.067. The 25th percentile, median and 75th percentile EQ-5D preference values were estimated. For those with Crohn's disease in remission these were 0.70, 0.80 and 1.00, while for those with mild disease they were 0.50, 0.72 and 0.80 and for those with moderate to severe disease they were 0.50, 0.60 and 0.70. For those with ulcerative colitis in remission these were 0.80, 1.00 and 1.00, while for those with mild disease they were 0.50, 0.72 and 0.80 and for those with moderate to severe disease they were 0.50, 0.50 and 0.70.

Bernklev et al administered the SF-36 among 166 Norwegian patients with Crohn's disease and 348 Norwegian patients with ulcerative colitis.[110] All patients with IBD or possible IBD in four areas of south eastern Norway had been identified 5 years previously. At the 5 year follow up 200 patients with Crohn's disease and 454 patients with ulcerative colitis remained diagnosed with IBD, with 166 and 348 of these respectively giving their consent to participate in the study. Patients with Crohn's disease had lower mean scores in all dimensions compared to patients with ulcerative colitis, but the paper does not appear to report whether these were significantly different or not. Both patients with Crohn's disease and patients with ulcerative colitis had significantly lower mean scores in all dimensions when matched with a reference population. Similarly, splitting patients into those with no symptoms, those with mild symptoms and those with moderate or severe symptoms saw symptom severity being statistically significant across all dimensions among both patients with Crohn's disease and patients with ulcerative colitis.

Bassi et al, available only in abstract, conducted face to face interviews with 120 IBD outpatients and 9 IBD inpatients, directly measuring quality of life using TTO, the VAS and the EQ-5D.[122] The average utility scores for Crohn's disease and ulcerative colitis were 0.84 and 0.89 using TTO, 0.62 and 0.70 using the VAS and 0.71 and 0.77 using the EQ-5D. For the TTO utilities, disease severity showed a significant negative correlation: -0.37 for Crohn's disease when measured by the Harvey Bradshaw index and -0.42 for ulcerative colitis when measured by the Simple Colitis Activity Index.

Crohn's disease: Quality of life studies

Arseneau et al, within the context of an assessment of the cost effectiveness of infliximab for Crohn's disease perianal fistulae in the United States, undertook a time trade off exercise among 32 Crohn's disease patients, 17 of whom were fistulising or had a history of fistulising and 15 who did not, and 20 health members of the general public.[123] For reasons that are unclear, the utilities for health states were also differentiated by whether a patient was receiving infliximab or was receiving 6MP/metronidazole therapy. This resulted in the following values.

Table 39. Arseneau et al (2001) Crohn's disease TTO utilities

	CD patients	Healthy individuals
Infliximab		
Fistula	0.73	0.77
Improved fistula	0.85	0.91
Perianal abscess	0.62	0.72
6MP		
Fistula	0.69	0.75
Improved fistula	0.81	0.88
Pancreatitis+fistula	0.47	
Pancreatitis	0.57	
Parenthesias+fistula	0.66	0.68
Parenthesias	0.75	0.84

Note that in the above the HRQoL values for pancreatitis health states are as per the footnote to Table 2 of Arseneau et al, the values reported in table 2 of Arseneau et al assuming that patients only spent one quarter of their time with pancreatitis. These values appear to relate to the healthy respondents, but this is not entirely clear from the text. While the absolute values vary, the differences in the HRQoL values for those on treatment with fistula and improved fistula are reasonably consistent at between 0.12 and 0.14. The difference between pancreatitis with and without fistula of 0.10 was similar to the 0.09 difference between parenthesias with and without fistula among Crohn's disease patients, but the corresponding 0.16 difference among healthy respondents was that bit larger.

Buxton et al[h] explored the possibility of mapping from the Inflammatory Bowel Disease Questionnaire (IBDQ) and from the Crohn's Disease Activity Index (CDAI) to utilities.[124] The data set consisted of paired contemporaneous observations from patients with moderate to severe Crohn's disease who participated in either of two natalizumab trials, with over 3,000 observations. Demographic data was not presented. Both the SF-36 and the EQ-5D were considered, with these being transformed onto utilities using the SF-6D and the UK social tariff respectively. The mean SF-6D utility was 0.68, while the mean EQ-5D utility was 0.70. The paper derives a mapping function for the SF-6D that is non-linear in the IBDQ, but the preferred mapping function of the EQ-5D utility is linear in the IBDQ: 0.03043+0.0043IBDQ, with an R^2 of 0.45.

[h] Sponsored by Elan Pharmaceuticals

Benedini et al[i] applied the EQ-5D among 162 Italian patients with active Crohn's disease and a CDAI score of more than 150 at baseline, with an additional 3 six monthly follow up visits.[125] The mean baseline EQ-5D score was 0.558, with this showing a gradual improvement over the follow up visits to 0.682, 0.728 and 0.739. The valuation method for the EQ-5D is unclear, with the paper referencing the UK social tariff but stating that the values fall on the interval 0 to 1.

Casellas et al measured the quality of life among 49 Spanish patients receiving infliximab and in remission.[126] The number in remission fell to 42 at 12 months, 32 at 24 months, 13 at 36 months and 13 at 48 months. Casellas et al report the 25th percentile, the median and the 75th percentile of the EQ-5D among the patients in remission, using the preference set of the Spanish EuroQol. At baseline these were 0.8, 1.0 and 1.0, with the median among those in remission remaining at 1.0 over the period of the study and the 25th percentile never dropping below 0.8.

Gregor et al recruited 180 inpatients and outpatients with Crohn's disease from a single Canadian tertiary centre in order to evaluate quality of life using time trade off (TTO), standard gamble (SG) and the visual analogue scale (VAS).[127] Follow-up data from a second visit 8 weeks later was obtained from 164 of these patients. Patients were ineligible if they required imminent surgical treatment, had a significant comorbidity, had had surgery in the last four weeks or were not "judged by the investigators to comprehend the choices being offered by the HRQoL questionnaires".

Patients were divided into four groups:

- Chronically active therapy resistant: treatment with prednisone at a dose of ≥10 mg daily, continuous methotrexate of purine antimetabolites for a minimum of six months and a CDAI score of ≥150. 52 patients of whom 62% were women, and a mean age of 35.
- Chronically active therapy responsive: treatment with prednisone at a dose of ≥10 mg daily, continuous methotrexate of purine antimetabolites for a minimum of six months and a CDAI score of <150. 34 patients of whom 53% were women, and a mean age of 31.
- Acute disease exacerbation: a recent flare in activity with a CDAI score ≥150, no steroid or immunosuppressive drug therapy in the 12 weeks preceding the flare and the initiation of prednisone or 5-aminosalicylic acid treatment. 45 patients of whom 49% were women, and a mean age of 34.
- Remission: a CDAI score of <150 for a minimum of 6 months and no systemic glucocorticoid or immune-suppressive drug therapy: 49 patients of whom 59% were women, and a mean age of 37.

[i] Sponsored by Merck, Sharp and Dohme

At baseline the TTO, the SG and the VAS mean values for these groups were 0.88, 0.74 and 0.61 for the chronically active therapy resistant, 0.98, 0.86 and 0.82 for the chronically active therapy responsive, 0.89, 0.77 and 0.60 for the acute disease exacerbation, 0.96, 0.88 and 0.84 for remission and 0.92, 0.81 and 0.71 across all patients.

Three hypothetical disease states were outlined:

- Mild Crohn's disease: Four or fewer bowel movements per day associated with occasional abdominal pain, only occasionally absent from school of work because of illness, and rarely tired or having disturbed sleep.
- Moderate Crohn's disease: More than four but fewer than eight bowel movements per day associated with tolerable abdominal pain and occasional blood, tiredness most days, frequent frustration and concern about the side effects of medication, and frequent absences from school or work because of illness.
- Severe Crohn's disease: More than eight bowel movements per day, frequent abdominal pain and bloody stools, always tired with difficulty sleeping, depressed and frustrated and worries about the need for surgery and the side effects of medication.

The mean results were broadly consistent between the first assessment and the follow-up assessment, with the mean TTOs being 0.95 and 0.96 for mild disease, 0.88 and 0.88 for moderate disease and 0.73 and 0.71 for severe disease. The mean SGs were 0.81 and 0.82 for mild disease, 0.72 and 0.73 for moderate disease and 0.50 and 0.54 for severe disease. The mean VAS scores were 0.80 and 0.82 for mild disease, 0.57 and 0.61 for moderate disease and 0.27 and 0.31 for severe disease. Within these results, though the absolute values for the TTO lie above those of the SG the net HRQoL changes from moving from mild to moderate disease, 0.07 to 0.09, and from moderate to severe disease, 0.15 to 0.22, are reasonably aligned between the TTO and the SG. The net changes estimated using the VAS are somewhat different: from moving from mild to moderate disease, 0.21 to 0.23, and from moderate to severe disease, 0.30.

Gibson et al[j] surveyed 143 patient with the AQoL questionnaire, recruited from five Australian outpatient clinics.[128] Patients had had their diagnosis of Crohn's disease confirmed by a specialist physician on standard clinical, radiological, endoscopic and histopathalogical criteria. Patients with significant comorbidities were excluded. 110 patients were without fistulae, while 23 had fistulae. The overall mean CDAI of 171 was slightly lower at 169 in those without fistulae and higher at 177 in those with fistulae but this difference was not significant. Those without fistulae were roughly equally balanced between ileal, ileocolonic and colonic while 64% of those with fistulae were colonic. The

[j] Sponsored by Schering-Plough

AQoL website outlines that the AQoL utilities scoring system is based upon TTO, though the EAG has not explored this in any depth. Among those without fistulae the average HRQoL was 0.646 compared to 0.606 for those with fistulae. For those without fistulae the average HRQoL was: 0.766 for those with a CDAI of less than 150; 0.680 for those with a CDAI of between 150 and 219; and, 0.450 for those with a CDAI of more than 220. Relating the HRQoL to the CDAI score there was a broadly negative relationship, though this showed quite a wide dispersion of points around the regression line of HRQoL = 0.8198 – 0.00107 *CDAI and the R^2 was only 0.27.

While of relatively limited usefulness for cost effectiveness modelling purposes, Hill et al report quality of life values among 41 Australian Crohn's patients who were paediatric at diagnosis.[129] Quality of life was measured using the IMPACT III questionnaire, composed of 35 questions each of which was scored on a 1 to 5 likert scale giving a range of possible values from 35 to 205 with a higher score being taken to indicate a better quality of life. These were further related to the PCDAI, with patients being grouped into remission with a PCDAI $\leq$ 10, mild disease with a PCDAI 11 - 29, or moderate to severe disease with a PCDAI $\geq$ 30. A multivariate analysis found QoL as measured by the IMPACT III questionnaire to be significantly affected by the PCDAI. Age, gender, disease duration and whether diagnosis was within 6 months were not found to be significant. Whether patients were receiving treatment, either drug or enteral nutrition, was of borderline significance (p=0.07) as was whether the patient was growth impaired as measure by the height z score (p=0.06).

Ulcerative Colitis: Quality of life studies

Connolly et al[k] analysed EQ-5D data from a study of Western European patients with mild to moderately active ulcerative colitis scoring between 3 and 8 points on the UCDAI. This compared oral mesalazine plus a daily mesalazine enema, n=71,with oral mesalazine plus a daily placebo enema, n=56, over a four week period, with an additional four week follow up period with no enemas.[130] The proportion of women in the mesalazine enema was 38% compared to 43% in the placebo enema arm, and the median ages were 42 years and 47 years respectively. The EQ-5D was administered at baseline, week 2, week 4 and week 8. The paper does not appear to report what value set was used to convert the EQ-5D to utility scores. At baseline the mean EQ-5D index values were 0.778 in the mesalazine enema arm and 0.762 in the placebo enema arm. These showed continuous improvement over the study period, including between week 4 and week 8 when the enemas had been discontinued, reaching 0.914 and 0.862 at week 8.

[k] Sponsored by Ferring Pharmaceuticals

Bryan et al in the ERG report for the STA of infliximab at a dose of 5mg/kg for the treatment of acute exacerbations of ulcerative colitis summarise the HRQoL data within the manufacturer submission.[131] This mainly relied upon the HoDAR study that measured the EQ-5D among 171 Welsh ulcerative colitis patients. Additional data for the HRQoL for surgery with complications health state of the submitted model was drawn from Arseneau et al which applied the TTO among 48 US patients with ulcerative colitis.[132] The ERG report tabulated these as below.

Table 40. Utility estimates associated with health states (from Bryan et al 2008)

	Arsenau TTO		HoDAR EQ-5D	
	Mean	s.d.	Mean	s.d.
Remission	0.79	0.24	0.88	0.14
Active ulcerative colitis	0.32	0.31	0.42	0.32
Surgical remission	0.63	0.30	0.60	0.38
Surgical complications	0.49	0.32		

Note that Feagan et al[l] used the IBDQ in a study of infliximab treatment for patients with moderate to severely active ulcerative colitis disease at doses of 5mg/kg and 10mg/kg with a placebo control arm.[133] The average IBDQ scores at baseline were 125, 130 and 124 respectively. Applying the IBDQ to HRQoL mapping function derived by Buxton et al for Crohn's disease to these mean scores results in HRQoL values of 0.568, 0.589 and 0.564. At week 8 the mean improvements in the IBDQ were 40, 36 and 21 respectively which would translate into HRQoL gains of 0.202, 0.185 and 0.121. These were broadly maintained to week 30. Feagan et al also noted mean improvements in the IBDQ for those with mucosal healing of 48 at week 8 and 58 at week 30, which translate into HRQoL gains of 0.237 and 0.280. For those without mucosal healing the corresponding IBDQ improvements were only 16 and 7, which translate into HRQoL gains of 0.099 and 0.061.

Waljee et al used time trade off to measure the quality of life and perceived quality of life with and without colectomy among US ulcerative patients recruited from primary care without a colectomy and ulcerative patients post-colectomy.[134] Unfortunately, throughout their paper Waljee et al only report the median values, though this is mitigated by the 25th percentiles and the 75th percentiles also being reported. For current purposes, the more interesting results are the quality of life values recorded among patients without a colectomy living with chronic mild (n=55), moderate (n=47) and severe (n=48) ulcerative colitis. The medians (interquartile range) for these were 0.96 (0.91-1.00), 0.94 (0.86-0.98) and 0.96 (0.88-0.99) respectively, while across the group as a whole they were 0.96 (0.89-0.99).

[l] Sponsored by Centocor Inc.

Poole et al[m] used trial data from the PINCE clinical trials to map between the UCDAI score of patients and the individual dimensions of the EQ-5D with these subsequently being mapped to utilities, presumably using the EQ-5D UK social tariff though this does not appear to be stated.[135] The observed EQ-5D utilities were compared with those estimated for both the PINCE trial and the separate PODIUM trial. For those in remission the mean utilities for PINCE observed, PINCE estimated and PODIUM estimated were 0.944, 0.939 and 0.940. For those with mild/moderate disease the mean utilities were 0.811, 0.801 and 0.775. For those in severe relapse the mean utilities were 0.700, 0.630 and 0.660. The reasons for the estimated utilities falling below the observed utilities for mild/moderate disease and for severe relapse is not clear.

Colonoscopy patient impacts and adverse events.

Baudet et al followed up 1,126 randomly selected Spanish colonoscopy patients, 78% of whom received sedation for the colonoscopy.[136] Sedation was on request, not randomly allocated. There were two episodes of bleeding, both of which followed the removal of very large polyps. There were no perforations. Early adverse events of bradychardia and hypoxia rates were 7.2% and 4.6% in the sedated group, compared to 3.2% and 1.2% in the non-sedated group, while tachycardia was less frequent in the sedated group at 2.5% compared to 9.2%. Nausea and vomiting occurred at an average 5.6% across the groups, while abdominal pain during with the procedure was less in the sedated group at 5.1% compared to 47.8% among the non-sedated group.

Patients were followed up by telephone interview 30 days after their colonoscopy. Abdominal pain occurred on average across 7.2% of those responding, though was lower at 1.9% among the sedated than the 29.7% among the non-sedated. Abdominal distension and bloating was also relatively common at 4.9%. Rectal bleeding occurred among 2.4% of patients. Baudet et al conclude that minor complications of colonoscopy are reasonably common. But it is unclear whether the reported events at 30 days were necessarily due to the colonoscopy or could also be linked to the condition under investigation.

Also of note is that sedation reduced the frequency of some adverse events during colonoscopy, including pain and discomfort, and allowed more extensive investigations, such as intubation of the caecum.

De Jonge et al followed up 1,144 Dutch colonoscopy patients by telephone interview. Major events were defined as those requiring hospital intervention.[137] For the major events that were definitely

[m] Sponsored by Ferring Pharmaceuticals

procedure related in the 30 days follow up period, 0.36% required hospitalisation due to rectal bleeding, while 0.18% required hospitalisation due to abdominal discomfort, while dizziness, perforation and angina pectoris each occurred in an additional 0.09% of patients. Only 3% of patients had major events. However 41% had minor adverse events. Those that were definitely procedure related in the 30 days follow up period were: abdominal discomfort in 17% of patients, rectal blood loss in 5.6%, and a change in bowel habit in 5.4%.

Dominitz et al undertook a time trade off study to investigate the amount of survival people would be willing to sacrifice to avoid 5 yearly screening with sigmoidoscopy or colonoscopy.[138] Four patient groups were involved: those with no experience of screening, those undergoing screening with sigmoidoscopy, those undergoing screening with colonoscopy, and those with colorectal cancer. Those with no experience of screening were willing to trade-off reasonable median amounts of time to avoid sigmoidoscopy and more time to avoid colonoscopy. Those screened with sigmoidoscopy were not willing to trade off any time to avoid sigmoidoscopy when measured at the median, but were willing to trade off some time to avoid colonoscopy. Those screened with colonoscopy and those with colorectal cancer were not willing to trade off any time to avoid sigmoidoscopy or to avoid colonoscopy when measured at the median. While there might be a degree of patient choice among those being screened by sigmoidoscopy and among those being screened by colonoscopy, the results would seem to suggest that the anticipation of the procedures may be worse than the reality.

Niv et al assessed quality of life using the SF-36 both pre, immediately post and 30 days after colonoscopy among 100 Israeli patients.[139] There were no significant changes before and immediately after the colonoscopy in any of the SF-36 parameters with all the scores having similar scores pre and post procedure. Similarly, scores were also similar at the one month point though there was a decrease noted in the physical functioning score. This applied among the non-IBD patients and not among the IBD patients, which might be suggestive of it being condition related rather than being procedure related.

Spiegel et al retrospectively evaluated 458 US patients with IBS using the SF-36, to examine whether having had a previous colonoscopy affected quality of life.[140] Controlling for potential confounding variables, Spiegel et al found no relationship between having had a colonoscopy and quality of life. They conclude that there was no evidence that the reassurance provided by a negative colonoscopy improved the quality of life of IBS patients.

Warren et al undertook a retrospective analysis of a random sample of 5% of Medicare beneficiaries aged 66 to 95 who had undergone a colonoscopy (n=53,220), matching these with controls in order to estimate whether colonoscopy raised event rates within 30 days of the colonoscopy.[141] Patients were

matched by date of birth, race, sex, state and a comorbidity score. Adjusting for covariates, they found that diagnostic colonoscopies were associated with a 0.42% risk of a serious GI event compared to 0.18% for those with no colonoscopy, an 8.9% risk of other GI events compared to 5.7% for those with no colonoscopy, but the same risks of cardiovascular events. But it remains unclear to what extent the patient matching would have controlled for the patient group having colonoscopies being inherently more likely to have GI conditions which would themselves lead to other GI events, without these being necessarily related to the colonoscopy.

Levin et al undertook a retrospective analysis of the medical records of 16,318 patients who had undergone a colonoscopy between 1 January 1994 and 16 July 2002 within the Kaiser Permanente health care system of Northern California to determine rates of serious complications within 30 days of the procedure.[142] Patients were eligible for inclusion if they were older than 40 years of age. Among the 5,235 procedures carried out without a biopsy none resulted in a serious bleed but 3 resulted in a perforation. Among the 11,083 procedures carried out without a biopsy 53 resulted in a serious bleed and 12 resulted in a perforation.

The economic modelling for CG118: Colorectal cancer – screening with colonoscopy, assumes that people on surveillance have no complications caused by colonoscopy such as perforations or bleeding.[143] This is probably due to the rarity of these events.

In contrast, the ScHARR Report to the English Bowel Cancer Screening Working Group estimates rates of bleeds and of perforation for colonoscopy and flexible sigmoidoscopy, and the mortality rates associated with the perforations.[144]

The UK flexible sigmoidoscopy trial reported 12 patients being admitted for bleeding following screening among the 40,764 people screened using it: a rate of 0.0295%[145]. 9 of the 2,051 patients undergoing colonoscopy with polypectomy were re-admitted to hospital with bleeding: a rate of 0.4390%.

The UK flexible sigmoidoscopy trial, as reported in Atkin et al apparently suggested only 1 perforation among the 40,764 people screened using flexible sigmoidoscopy.[146] For colonoscopy with polypectomy, Atkin et al reported 4 perforations out of 2,377 colonoscopies performed: a rate of 0.168%. The ScHARR report halved this rate for colonoscopies without polypectomy.

The probability of dying following a perforation was drawn from the study by Gatto et al, which randomly sampled 5% of Medicare beneficiaries within certain regions of the US.[147] These figures

need to be treated with caution, as all patients were over 65 years of age, but from a total of 108 perforations recorded 6 patients died: 5.56%.

Table 41. Probability of dying following a perforation (from Gatto et al 2003)

Event	No polypectomy	With polypectomy
Colonoscopy bleed	0.4390%	
Colonoscopy perforation	0.0800%	0.1680%
Colonoscopy mortality given perforation	5.2%	
Sigmoidoscopy bleed	0.0295%	
Sigmoidoscopy perforation	0.0025%	
Sigmoidoscopy mortality given perforation	6.4%	

Bleeds were assume to require one night as an inpatient, while treating a perforation was assumed to require major surgery. Based upon 2011-12 NHS reference costs bleeds could be costed at the non-elective inpatient stay FZ38F: £561 [IQR: £339 to £783], while the cost most in line with the ScHARR study for perforations appears to be the non-elective inpatient stay FZ77A: £5,360 [IQR: £3,368 to £6,390].

Cost utility modelling

Summary of modelling approach

The modelling required for a full cost utility modelling exercise is complicated by there being at least three main conditions under consideration, IBS, Crohn's disease and ulcerative colitis, and a range of other considerations when paediatric IBD versus non-IBD are compared, because IBS, while still the commonest non-IBD diagnosis, is less common than in adults. Modelling induction and maintenance subsequent to diagnosis for these three conditions is quite involved. As a consequence, an initial consideration of the truncated quality of life impacts for time spent as false negatives is presented which can be considered alongside the costs of the initial test sequences and likely periods of time spent as false negatives. This quality of life impact is restricted to the direct detrimental quality of life impacts from not being correctly treated and not entering remission, this being limited by the time spent being incorrectly treated prior to representing for testing. EAG expert opinion suggests that 12 weeks is a reasonable base case assumption for the duration of false negatives being incorrectly treated prior to the possibility of IBD being re-considered.

But the truncated cost utility approach is formally incorrect, in that not all IBD patients when diagnosed with either Crohn's disease or ulcerative colitis will immediately enter remission after treatment and remain in remission thereafter. Moreover, achieving remission and maintaining it is not costless. As a consequence, it appears that there is a requirement for a full cost utility modelling exercise that takes into account the costs and benefits of induction therapy and maintenance therapy in both Crohn's disease and ulcerative colitis, bearing in mind the potential problems of false negatives (IBD missed).. This is the approach adopted by the EAG, but the EAG is of the opinion that the

outputs of a truncated analysis could provide a useful sense check to the results of the full cost utility modelling, particularly in the light of the latter's complexity and that its costs are an order of magnitude or more greater than the up-front testing costs.

The modelling for the full cost utility approach is eased by the modelling of induction and maintenance of remission for CG152: Crohn's disease: Management in adults, children and young people and the modelling for the draft clinical guideline for ulcerative colitis being available.[102,103] Both sets of models adopt a similar framework. Induction therapy with the aim of remission, but with subsequent induction therapies for those not achieving remission. Those achieving remission enter a maintenance of remission model, most patients being on treatment but a relatively small minority maintaining remission without active therapy. Remission can be lost, however, which leads to a further sequence of induction therapies. Note that the sequences of induction therapies in the initial induction therapy modelling and the sequences of induction therapies among those having lost remission in the maintenance of remission modelling therapy modelling differ, and even where the same therapy is involved it may have different clinical effectiveness estimates. All induction therapy sequences have as their final option surgery. This, in common with the modelling for the clinical guidelines, is assumed to achieve a permanent remission without the requirement for any further therapy, though this may be optimistic given the 10-year time horizon of the modelling.

Within this modelling, in common with the clinical guidelines' modelling, there is no explicit consideration of possible disease progression such as the development of fistula during the period of time spent being incorrectly treated as false negative or during periods of loss of remission. Were this to apply, it is likely that the relative importance of sensitivity over specificity would increase compared to the current modelling approach. We note that in the study by Shaol et al[83], two cases had fistulae at diagnosis.

The above has made little reference to the modelling of relief of symptoms in IBS patients. The approach adopted is broadly in line with that of the YHEC model, informed by the modelling for CG61: Diagnosis and management of irritable bowel syndrome in primary care.[35] This is simpler than the modelling of induction and maintenance in Crohn's disease and in ulcerative colitis. But given the assumed 100% specificity of colonoscopy meaning that there are no false positives at the end of the test sequence, the modelling of IBS and its treatment subsequent to diagnosis is of lesser importance within the overall cost utility modelling. It is in effect a common residual to all comparators. Its main impact is to determine the costs incurred among false negatives being incorrectly treated. Given this, the full cost utility model can be viewed as both an IBD vs IBS model and a reasonable IBD vs non-IBD model, provided that for the latter the costs among the false negatives are appropriately adjusted

to take into account any additional testing and treatments that may occur among the non-IBD patients, noting the lower proportion with IBS in children.

Two scenarios are modelled:

- Adult patients in primary care, with test accuracies for IBD versus IBS
- Paediatric patients in secondary care, with test accuracies for IBD versus non-IBD

Perspective, time horizon and discounting

The modelling adopts the NICE reference case perspective of patient benefits and NHS and PSS costs, over a 10 year time horizon for the base case with discounting of costs and benefits at an annual 3.5%.

Model structure

For reasons of space, the cost utility model is most simply presented as a set of interlinked models:

- The test model
- The induction and maintenance model among true +ve Crohn's disease patients
- The induction and maintenance model among true +ve ulcerative colitis patients
- The induction and maintenance model among true -ve IBS patients and false -ve IBD patients

The model structure for the initial testing sequence is as below.

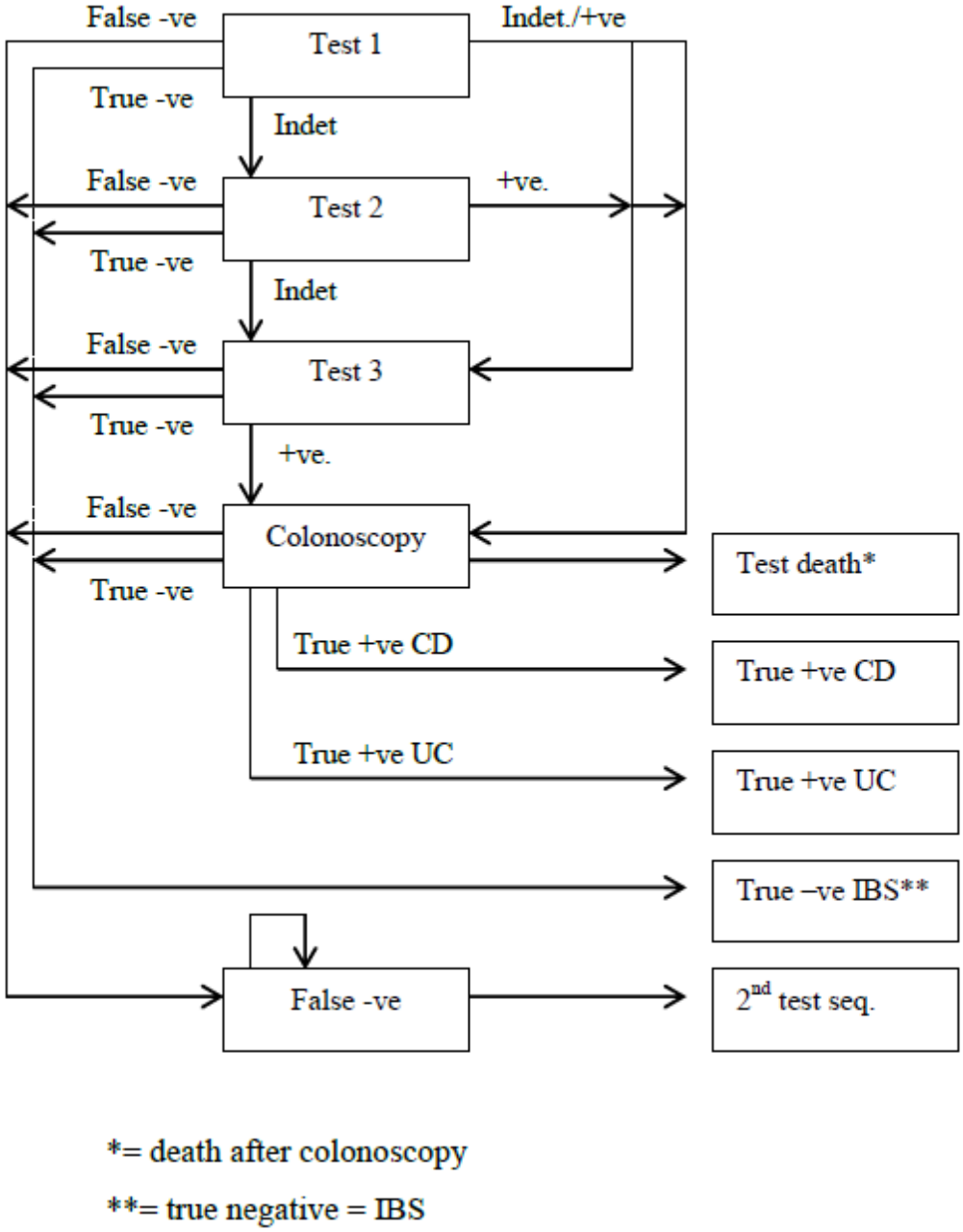


Figure 21. Model structure of initial test sequences

Due to the timing of testing and the possible delays between tests, all the models employ a weekly cycle. The delay between referral and colonoscopy is assumed to be 4 weeks and the time to retesting among those testing negative but not responding to IBS therapy is assumed to be 12 weeks, both estimates being based upon expert opinion. This may be optimistic as noted in the Introduction, because a sequence of unsuccessful treatments may be pursued for IBS, and so is explored in sensitivity analyses.

The above permits a range of test sequences to be compared. For instance, an initial POCT with a poor specificity could be followed by an ELISA test. Faecal calprotectin testing does not have to result in an immediate referral for colonoscopy for all positive results. In a similar vein, the model structure also permits the exploration of rates of indeterminate test results having a follow up test prior to any referral to colonoscopy. Due to a lack of data this latter option has not been explored in the analyses which follow.

The key assumption in all of the above is that all those who test positive, possible after a sequence of tests, receive a colonoscopy. The current modelling, as outlined below, assumes that referrals to secondary care result in colonoscopy. The model structure allows for referral to secondary care to result in assessment by a gastroenterologist, with only a proportion of those referred going on to colonoscopy. But this requires that the sensitivity and specificity of any gastroenterology assessment be estimated. A lack of data means that this option has not been considered. The sensitivity and specificity of an ELISA test could be seen as the closest available proxy for this.

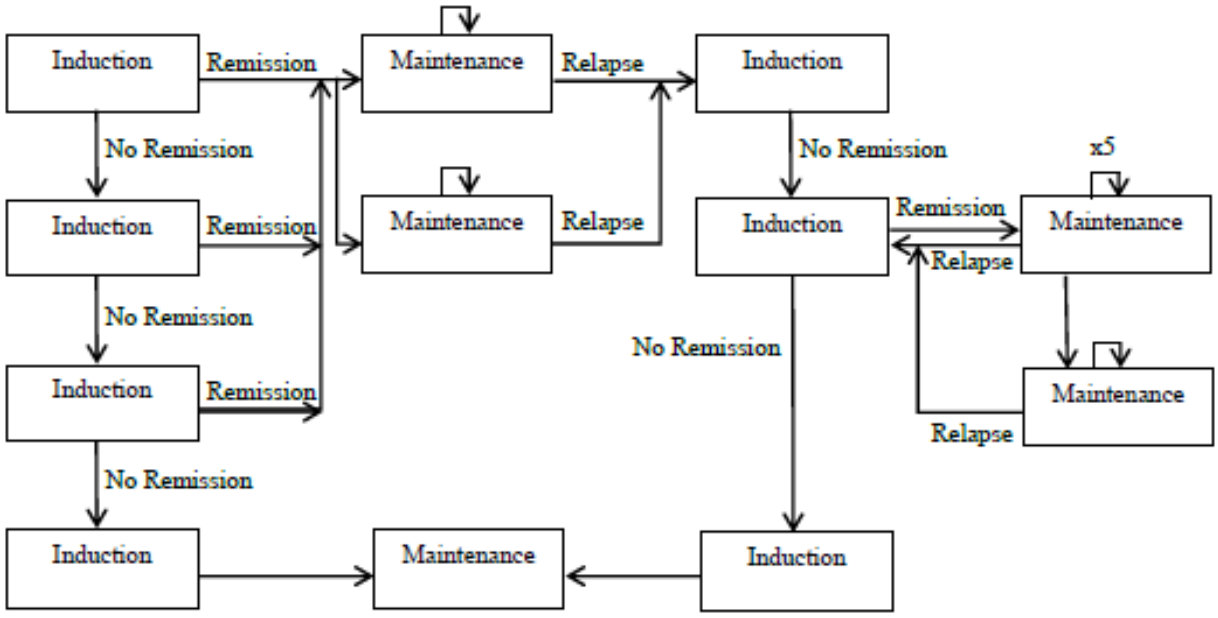


Figure 22. Model structure of Crohn's disease true positives

For the initial induction of remission in Crohn's disease, the most cost effective strategy within the modelling of CG152 was an 8 week course of prednisolone, followed by an 8 week course that adds azathioprine to prednisolone, followed by a 6 week course of anti-TNF. Within this, the more cost effective anti-TNF was adalimumab and this is applied within the base case modelling. This appears

to be broadly in line with TA187, though this envisages treatment with an anti-TNF for unresponsive disease for up to 12 months.

For the modelling of the maintenance of remission for Crohn's disease, this is again based upon the most cost effective strategy identified within the modelling of CG152. This assumes azathioprine as the maintenance therapy, followed by the same induction sequence as in the initial induction of remission modelling.

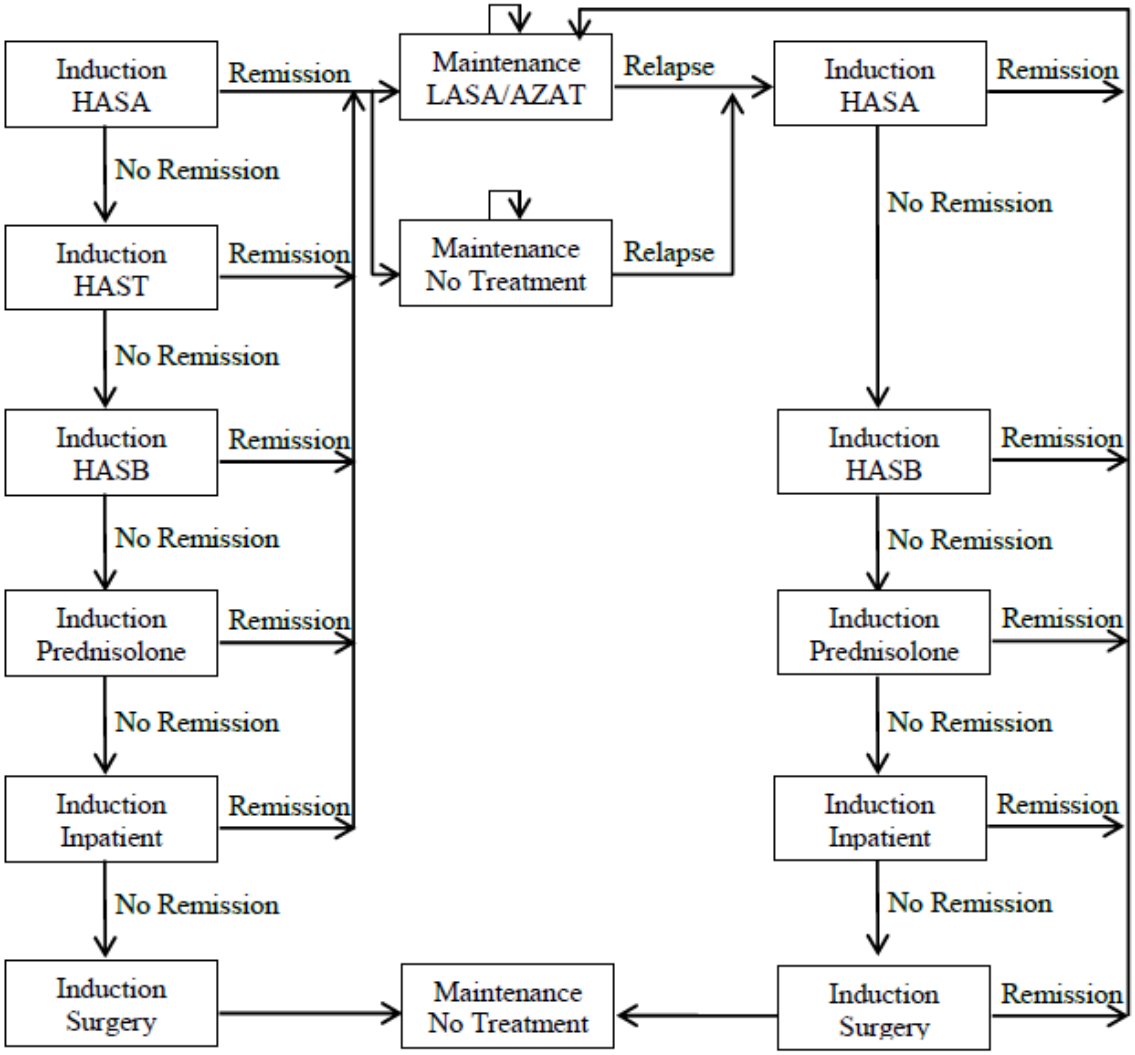


Figure 23. Model structure of ulcerative colitis true positives

In the above HASA is high dose ASA, LASA is low dose ASA HAST is high dose ASA with a topical ASA and HASB is high dose ASA with beclometasone. For diagrammatical simplicity, remission from inpatient therapy receives azathioprine maintenance therapy. All other patients receive LASA maintenance therapy if on active treatment.

For the modelling of induction of remission for ulcerative colitis, this is based upon the most cost effective strategies identified within the modelling for the draft ulcerative colitis guideline: strategy 10 of table 34 of appendix L: high dose ASA followed by addition of a topical ASA, followed by high dose ASA with beclometasone, followed by prednisolone.[103] This appears to be broadly in line with the recommendations of the draft clinical guideline. Whether induction of remission would initiate with the high dose ASA of strategy 10 or perhaps the low dose ASA of strategy 6 is a moot point. The net monetary benefits of the two strategies are similar: £8,513 and £8,323 at a willingness to pay of £20,000 per QALY, but the cost effectiveness of strategy 10 versus strategy 6 is estimated to be £2,818 per QALY with a likelihood of cost effectiveness of 47% compared to 18% for strategy 6. The current modelling adopts the sequence of strategy 10.

For the modelling of the maintenance of remission for ulcerative colitis, this is again based upon the most cost effective strategies identified within the modelling for the draft ulcerative colitis guideline: low dose ASA maintenance, followed by low dose ASA maintenance for any patients losing but then regaining remission, as in table 59 of appendix L.

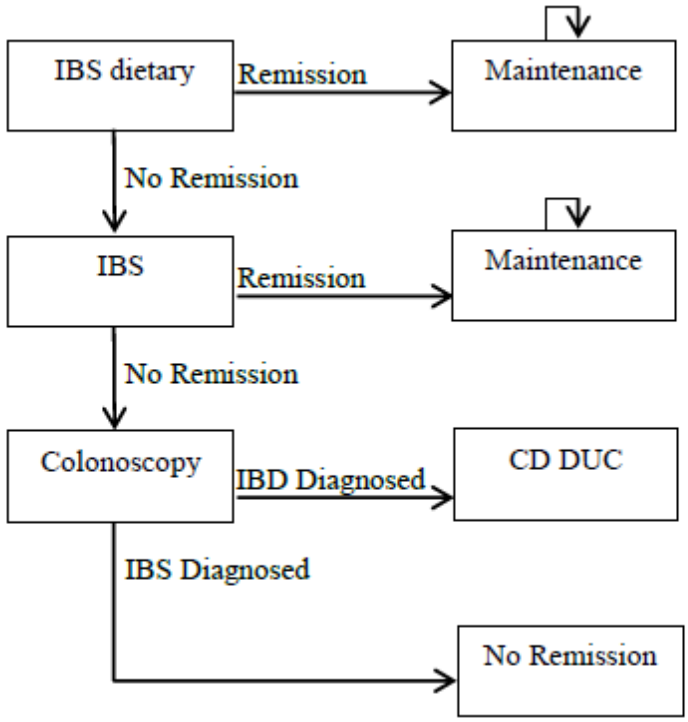


Figure 24. Model structure of IBS true negatives and IBD false negatives

The rate of response to the various treatments for IBS affects the total costs, in that those not responding to initial treatment with dietary advice followed by medication are assumed to be referred for a colonoscopy if they have not already had one. The economic review and modelling of the IBS clinical guideline suggests a 45% response rate for placebo as drawn from Mearin et al, which for the base case will be taken to be the response rate to dietary advice.[148] This is broadly in line with the 50% assumed in the YHEC report, which was based on expert opinion.

The IBS clinical guideline also outlines a range of medical therapies for IBS, with relative risks of response (compared to placebo) ranging from 1.32 for antispasmodics to 2.00 for anti-motility agents. As these can frequently be sequenced, this increases the overall response rates for medical therapies; e.g. sequencing in the antispasmodics modelling alone increases the estimated overall response rate to 78% in the modelling for CG61. In the light of this, the 5% colonoscopy referral rate for those not responding to initial treatment with dietary advice followed by medication of the YHEC report, based upon expert opinion, appears reasonable.

Note that for IBS patients who have already had a colonoscopy it is assumed that the colonoscopy is not repeated. There is some inconsistency of approach in this, in that the small proportion of false negatives who have previously had a colonoscopy are assumed to receive a colonoscopy. This can be justified upon grounds of presentation subsequent to incorrect treatment for IBS, but is not without objection.

The above has been applied in the modelling of IBD versus non-IBD diagnosis, the implicit assumption being that similar delay to representation and costs are incurred among false negatives. This may not be the case, and given the other conditions than IBS within non-IBD it may be that the average weekly cost for false negatives in the IBD versus non-IBD modelling should be higher than those for the IBD versus IBS modelling.

For the costs of IBS, Akehurst et al estimated a net additional annual cost of £188 [£123] compared to a control group of patients.[5] This suggests a weekly average of £9.29. For patients receiving medication for IBS a relatively minor additional £1.41has been included, based upon a simple average of the cost of generic mebevirine and the cost of the branded mebevirine, Colofac.

Primary care modelling: Base case test characteristics

The base case considers: the cost effectiveness of GP testing without faecal calprotectin being available; CalDetect at the 15µg/g cut-off as drawn from Otten et al[65]; and, ELISA at the 50µg/g cut-off as drawn from figure 6 of the clinical review. The base case using the lower 15µg/g cut-off of Otten et al may initially seem surprising, but the data for the 60µg/g cut-off of Otten et al suggests

only a slight gain in terms of a better specificity, 97.8% compared to 94.5%, but significant loss in terms of a worse sensitivity, 60.9% compared to 100.0%.

Table 42. Base case test characteristics

Test	GP no testing	CalDetect	ELISA RoC	Colonoscopy
Cut-off	..	15µg/g	50µg/g	..
Sensitivity	100.0%	100.0%	93.0%	95.0%
Specificity	78.8%	94.5%	94.0%	100.0%
Cost	Nil additional	£24.03	£22.79	£741.68[n]

The costs of Caldetect are based upon the list price for KST11005 PreventID CalDetect on the AlphaLabs website, coupled with 15 minutes of GP practice nurse time. ELISA testing is based upon an assumption of 40 patient samples per 96 well plate, costed at the list price on the AlphaLabs website, coupled with an average 11-12 minutes of staff time at grade 6/7 of the NHS terms and conditions of service handbook: Annex C Table 12. These staff costs have been proportionately increased for oncosts in line with the oncosts estimated for a hospital pharmacist within the PSSRU 2011 Unit costs of health and social care[o].

The resulting staff costs are very similar between all faecal calprotectin tests, and the main cost differences are due to the publicly available prices for the consumables. During this assessment it has been consistently noted by both suppliers and those using the tests within the NHS that most if not all tests are sold at a discount to the publicly available prices. These discounts will differ between tests, and possibly even between suppliers of a given test. There will be further geographic variation. Probably the best that the EAG can do is to consider a range of hypothetical discounts common across all tests, but given the base case results that follow there is little requirement for this.

The accuracy of colonoscopy is drawn from expert opinion.

Note that in the above the colonoscopy cost includes the cost of an outpatient gastroenterology visit at a cost of £164[p] and the costs of adverse events. Given the rarity of bleeds and perforations, despite the large cost associated with perforation these add very little to the overall costs of investigation: only around £12 to the cost of each colonoscopy.

Secondary care modelling: Base case test characteristics

[n] Weighted average of NHS reference cost OP and day case FZ51Z without biopsy, or FZ52Z with biopsy. Base case assumes 100% colonoscopy with biopsy. Sigmoidoscopy where included is costed as the weighted average of OP and day case FZ54Z without biopsy, or FZ55Z with biopsy. Also includes the cost of a gastroenterology OP appointment.

[o] Note that for the later scenario analyses a £28.27 cost of Quantum Blue is based upon the list price for Quantum Blue on the AlphaLabs website, the list price for an extraction kit on the BioHit website, coupled with an average 12-13 minutes of staff time at grade 6/7. Staff times for sample preparation for Quantum Blue and ELISA have been equalised.

[p] NHS reference cost: 301 Gastroenterology Consultant led First Face to face Non Admitted

The base case considers: the cost effectiveness ELISA at the 50µg/g cut-off as drawn from figure 13 of the clinical review; the cost effectiveness ELISA at the 100µg/g cut-off as drawn from figure 16 of the clinical review; and all patients being referred directly to colonoscopy. This results in the following test characteristics for the secondary care modelling.

Table Base case characteristics: secondary care

Test	ELISA	ELISA	Colonoscopy
Cut-off	50µg/g	100µg/g	..
Sensitivity	99.0%	94.0%	95.0%
Specificity	74.0%	82.0%	100.0%

Characterising uncertainty around the RoC curves

For the PSA, a possible and correct approach to characterising uncertainty around the central estimates drawn from the RoC curves is to draw a sufficient number of estimates from iterations of the WINBUGs code to characterise the distributions. This has the advantage of correlating the sensitivity and specificity that underlies the RoC curve estimates. But for current purposes this required a large number of iterations for their means to converge to the central estimates of the RoC curves. Given the size and complexity of the other modelling this would have led to each PSA requiring a very long time to run.

In the light of this a simpler approach has been adopted. The sensitivity and specificity, or rather their deviation from 100% accuracy, is simulated using the gamma distribution. Over 1,000 iterations this results in the same central estimates as figures 6, 13 and 16, but slight differences in the upper and lower confidence limits as outlined below. Any discrepancies appear minor, with the possible exception of the lower confidence interval limit for the specificity of figure 6, possibly due to this data being very heavily skewed compared to the other estimates. But it should be borne in mind that these simulations assume independence between each RoC's sensitivity and the specificity

Table 43. PSA simulated sensitivities and specificities for the ROC curves

			Lower CI		Upper CI	
		Central	Actual	Simulated	Actual	Simulated
Figure 6	Sensitivity	93%	83%	85%	97%	98%
	Specificity	94%	73%	76%	99%	100%
Figure 13	Sensitivity	99%	95%	95%	100%	100%
	Specificity	74%	59%	59%	86%	85%
Figure 16	Sensitivity	94%	86%	87%	98%	99%
	Specificity	82%	67%	68%	91%	92%

Baseline patient characteristics: Primary care

For the primary care adult population, the model adopts a baseline age for those presenting of 25 years as drawn from the Crohn's disease CG152 modelling, though this may be quite low for IBS patients.[102] In line with the Crohn's disease CG152 modelling, the female proportion is taken to be 50% for both Crohn's disease and ulcerative colitis. IBS appears to have a higher proportion of women presenting, the Brazier et al sample being 86% female though this estimate may be towards the upper end.[111] The base case adopts a 75% female proportion for IBS. Note that these estimates only affect the all population mortality risks. Since these are low during mid-adulthood for both women and men, the average age and proportion of women inputs have minimal impact upon results.

The base case 6.3% (7/111) prevalence of IBD is drawn from the Durham data while the 60% (539/904) prevalence of ulcerative colitis among IBD patients is drawn from Shivananda et al.[25]

Baseline patient characteristics: Secondary care

For the secondary care paediatric population, female proportions of 38% (35/91) for IBD patients and for 44% (44/99) non-IBD patients are drawn from Henderson et al.[26] An average age of 16 years is assumed, though as for the adult modelling this has minimal impact upon results.

The base case 48% (91/190) prevalence of IBD and the 75% (62/83) prevalence of Crohn's disease among IBD patients are drawn from Henderson et al.

HRQoL

While Konig et al provide quality of life estimates for both Crohn's disease and ulcerative colitis using the EQ-5D, their relevance is limited by the German mapping function being used.[116] Similarly, though Casellas et al also provide quality of life estimates for both Crohn's disease and ulcerative colitis using the EQ-5D, their relevance is limited by the Spanish mapping function being used.[120] Stark et al provide quality of life estimates for both Crohn's disease and ulcerative colitis using the EQ-5D and the UK social tariff.[118] Their sample sizes are also reasonably large: 270 with Crohn's disease and 253 with ulcerative colitis, though there may be some concerns around sample selection given that only a little over one third of the 1,447 originally contacted agreed to participate. Despite this, Stark et al appear to provide the most coherent set of utility values in line with the NICE reference case, the values of interest being 0.890 for remission and 0.610 for active disease for Crohn's disease patients and 0.910 for remission and 0.710 for active disease for ulcerative colitis patients. These imply utility decrements for active disease compared to remission of 0.280 for Crohn's disease patients and 0.200 for ulcerative colitis patients.

Of the Crohn's disease specific quality of life papers, Gregor et al is of the most interest.[127] Their TTO results suggest quality of life values of 0.955 for mild disease, 0.880 for moderate disease and 0.720 for severe disease: decrements from mild disease of 0.075 for moderate disease and 0.235 for severe disease. While the 0.235 decrement for mild to severe disease is in line with the 0.280 estimate of Stark et al, for current purposes the 0.075 decrement for mild to moderate disease might be the relevant estimate, or at least be applicable to a larger proportion of patients. This could suggest a somewhat lesser impact from false negatives being incorrectly treated than occurs with the Stark et al estimates, though note that within the Stark et al patient population only 0.4% of Crohn's disease patients had severe disease. CG152 uses the 0.280 decrement of Stark et al.

Of the ulcerative colitis specific quality of life papers, the reporting in Bryan et al of the HoDAR EQ-5D values of 0.880 for remission and for 0.420 active disease suggest quite a large decrement of 0.460.[131] This appears to be out of line with the other estimates that are available. The draft clinical guideline for ulcerative colitis uses the values of Poole et al of 0.940 for remission and 0.775 for mild to moderate disease, suggesting a decrement of 0.165.[135] This is slightly less than the decrement of 0.200 of Stark et al and would also suggest a lesser impact from false negatives being incorrectly treated than occurs with the Stark et al estimates, though note again that within the Stark et al patient population only 2.2% of ulcerative colitis patients had severe disease.

In the light of this, the base case will apply the quality of life decrements from remission to active disease of 0.280 for Crohn's disease and 0.200 for ulcerative colitis of Stark et al. But sensitivity analyses applying the quality of life decrements from mild to moderate disease of 0.075 for Crohn's disease as drawn from Gregor et al and of 0.165 as drawn from Poole et al will also be explored.[127,135]

It can be argued that the quality of life data for those with missed IBD who are being incorrectly treated for IBS may differ from that of patients with IBD who are not in remission but are being correctly treated for IBD. But in the absence of quality of life estimates for those with missed IBD who are being incorrectly treated for IBS, the best proxies are the quality of life estimates for those patients with IBD who are not in remission but are being correctly treated for IBD.

In this context it is important to bear in mind that there is also uncertainty around the average duration that false negatives will be incorrectly treated for IBS before representing and being further investigated due to IBS treatment not inducing remission. EAG expert opinion suggests that an average of 3 months is reasonable, with the main quality of life impacts being broadly proportionate to this duration. Given this, the total QALY decrements among false negatives during their period of

incorrect treatment for IBS can be presented for the quality of life decrements outlined above, coupled with a range of possible durations of incorrect treatment. For the base case 3 month duration, the QALY decrement is simply one quarter of the quality of life decrement.

Table 44. QALY decrements for different utility estimates and durations of false negatives

			Total QALY decrement from being a false negative for:					
Condition	Source	Decrement	1 month	2 months	3 months	4 months	5 months	6 months
Crohn's disease	Stark et al	0.280	0.023	0.047	0.070	0.093	0.117	0.140
Crohn's disease	Gregor et al	0.075	0.006	0.013	0.019	0.025	0.031	0.038
Ulcerative colitis	Stark et al	0.200	0.017	0.033	0.050	0.067	0.083	0.100
Ulcerative colitis	Poole et al	0.165	0.014	0.028	0.041	0.055	0.069	0.083

For Crohn's disease, retaining the estimates of Stark et al and moving from a 2 months' average duration to a 4 months' average duration doubles the overall QALY decrement as would be anticipated. While it can be argued that the sensitivity analysis using the decrement of Gregor et al is more speculative as it is experimental data, applying the decrement of Gregor et al within the context of a 2 months' average duration results in an overall decrement of only 0.013 QALYs compared to 0.093 QALYs when applying the decrement of Stark et al within the context of a 4 months' average duration: over a sevenfold difference.

For ulcerative colitis, applying the decrement of Poole et al within the context of a 2 months' average duration results in an overall decrement of 0.028 QALYs compared to 0.067 QALYs when applying the decrement of Stark et al within the context of a 4 months' average duration: between two and three times the amount.

These QALY decrements will be qualified by the prevalence of IBD in the presenting patient population, and the proportion of these who are modelled as being diagnosed as false negatives. For instance, an IBD prevalence of 5% coupled with a sensitivity of 90% results in only 0.5% of the total patient population being diagnosed as false negatives.

The above underlines that however complicated the full cost utility modelling is, the QALY decrements among false negatives will be dependent upon:

- the source of the quality of life values
- the assumed duration of patients remaining as false negatives
- the prevalence of IBD in the presenting population
- the sensitivity of the tests under consideration

Due to the low prevalence of IBD in the primary care population and the quite high sensitivities of the various tests the total QALY decrements among false negatives are likely to be quite small. Results may be mainly driven by the direct up-front test costs, including the costs of colonoscopies. This may also cause the adverse events and associated mortality from colonoscopy to come more to the fore, despite the assumed mortality rate also being very low.

The utility decrements for IBS are less important for current modelling purposes, given the 100% specificity assumed for colonoscopy meaning that there are no false positives by the end of the initial test sequence. For the base case, the 0.071 increment for response to treatment estimated within CG61 will be applied. The 0.662 baseline HRQoL that this increment is applied to is taken from Brazier et al.[111] A sensitivity analysis using the EQ-5D values of Spiegel et al can also be considered; 0.780 for response to treatment and 0.730 for no response to treatment, but recall that the mapping function employed by Spiegel et al is not clear.[3] Note that the baseline HRQoL value for IBS will also have an impact due to the small mortality rate associated with colonoscopy, with this impact enduring for the 10 year time horizon of the model.

Other model inputs

Given the extent of the downstream modelling, the full set of model inputs is presented in appendix 7, coupled with their treatment within the PSA.

Primary care modelling: Sensitivity and scenario analyses

The prevalence of IBD within the presenting patient population determines the relative importance of sensitivity and specificity. This is explored through sensitivity analyses that vary this from 5% to 25% in 5% increments.

A scenario analysis that speculates that faecal calprotectin testing might be used in a wider patient group than would be referred in the absence of faecal calprotectin testing is then presented.

Further sensitivity analyses are then presented which:

- Vary the time to representation among false negatives from the base case 12 weeks to 8 weeks,16 weeks and 24 weeks.
- Change the source of utility estimates from Stark et al and CG61 to Gergor et al , Poole et al and Spiegel et al.
- Remove the cost of the gastroenterology outpatient appointment from the cost of colonoscopy. (This was the approach used in the YHEC 2010 report and is included here to allow comparison with their figures. It also provides a sensitivity analysis around the cost of

referral and colonoscopy. Usual UK practice would be to refer for a gastroenterology opinion that would lead to colonoscopy, but in some countries, direct referral to colonoscopy appears to apply.)

- Vary the assumed non-response to medication among IBS patients from the base case 5% to 0% and 10%. (This applies to those in whom dietary advice has failed.)
- Remove the mortality associated with colonoscopy.

The clinical effectiveness section also presents a range of sources that include estimates of the accuracy of faecal calprotectin testing at various cut-offs. These are considered within individual scenario analyses, with the different cut-offs being directly compared, though these estimates are not integrated into the primary care base case.

Primary care modelling: Other estimates of test characteristics

Additional effectiveness estimates for further analyses are drawn from: Otten et al[65] for CalDetect at the 60µg/g cut-off; from Hessells et al[92] for CalDetect at the 15µg/g and 60µg/g cut-offs; from Hessells et al for Quantum Blue at the 30µg/g, 40µg/g, 50µg/g and 60µg/g cut-offs; and, from Basumani et al[75] for ELISA at the 50µg/g, 100µg/g and 150µg/g cut-offs. The central estimates from Otten et al are as below.

Table 45. Otten et al (2008) CalDetect accuracy

Test	CalDetect	CalDetect
Cut-off	15µg/g	60µg/g
Sensitivity	100.0%	60.9%
Specificity	94.5%	97.8%

In order to characterise the sensitivities and specificities of Hessells et al for the probabilistic modelling, the numbers of true positives, false negatives, true negatives and false positives has to be calculated. Unfortunately, Hessells et al only report the overall sample size, the numbers of correct diagnoses and the sensitivities and specificities. Given this, on the basis of a sample size of 85 the EAG has calculated the number with IBD to be 23, which implies the following numbers of true positives, false negatives, true negatives and false positives. These in turn imply sensitivities and specificities. In the main these correspond with those of Hessells et al, though there is some very minor disagreement of the order of 1% for a few of the percentages.

Table 46. Hessells et al (2012) POCT's accuracy

Test	Quantum Blue				CalDetect	
Cut-off	30µg/g	40µg/g	50µg/g	60µg/g	15µg/g	60µg/g
N with	23	23	23	23	23	23
TP	22	21	20	18	22	20
FN	1	2	3	5	1	3
Implied sensitivity	96%	91%	87%	78%	96%	87%
Hessells et al Table 1	96%	92%	88%	79%	96%	88%
N without	62	62	62	62	62	62
TN	43	52	52	54	33	46
FP	19	10	10	8	29	16
Implied specificity	69%	84%	84%	87%	53%	74%
Hessells et al Table 1	69%	84%	84%	87%	53%	74%

Table 47. Basumani et al (2012) ELISA's accuracy

Test	ELISA	ELISA	ELISA
Cut-off	50µg/g	100µg/g	150µg/g
Sensitivity	[redacted]	[redacted]	[redacted]
Specificity	[redacted]	[redacted]	[redacted]

Secondary care modelling: Sensitivity and scenario analyses

Sensitivity analyses are presented which:

- Vary the prevalence of IBD within the presenting patient population from the base case 48% to 40% and 60%.
- Vary the time to re-presentation among false negatives from the base case 12 weeks to 8 weeks and 16 weeks.
- Change the source of utility estimates from Stark et al and CG61 to Gregor et al, Poole et al and Spiegel et al.
- Doubling the annualised net cost amongst false negatives from £188 to £376
- Remove the mortality associated with colonoscopy.

Base case results: Primary care

For the primary care base case, the patient numbers receiving the initial test and being referred for colonoscopy are as below.

Table 48. Primary Care: Base case results of initial test sequence

	GP			CalDetect 15µg/g			ELISA		
	1st test	Colon.	Final	1st test	Colon.	Final	1st test	Colon.	Final
IBD tested	6.3%	6.3%		6.3%	6.3%		6.3%	5.9%	
True +ve	6.3%	6.0%	6.0%	6.3%	6.0%	6.0%	5.9%	5.6%	5.6%
False -ve	0.0%	0.3%	0.3%	0.0%	0.3%	0.3%	0.4%	0.3%	0.7%
IBS tested	93.7%	19.8%		93.7%	5.1%		93.7%	5.6%	
True -ve	73.9%	19.8%	93.7%	88.5%	5.1%	93.7%	88.1%	5.6%	93.7%
False +ve	19.8%	0.0%	0.0%	5.1%	0.0%	0.0%	5.6%	0.0%	0.0%

Note that the above relates to the initial test sequence of; e.g. CalDetect 15µg/g followed by colonoscopy. Within this test sequence, among those with IBD the initial Caldetect test identifies all 6.3% of patients with IBD as true +ves. The colonoscopy subsequent to this identifies 6.0% of the 6.3% referred by Caldetect as true +ves, due to its 95% sensitivity. Among those with IBS, the initial Caldetect test identifies 5.1% of the 93.7% of patients with IBD as false +ves. These are referred on, with the subsequent colonoscopy identifying all these as true –ves due to its 100% specificity. As a consequence, though the initial test referred on a proportion of false +ves these are all eliminated by the colonoscopy and at the end of the test sequence there are no false +ves.

Immediately apparent from the above is that GP opinion (without calprotectin) results in a somewhat larger number of false +ves being referred for unnecessary colonoscopies: 19.8% of the total patient population or 21.2% of those with IBS, as would be anticipated from the 78.8% specificity. CalDetect 15µg/g is somewhat better: 5.1% of the total patient population or 5.4% of those with IBS, as would be anticipated from the 94.5% specificity. The ELISA test, while perhaps marginally cheaper than the CalDetect test, is estimated to have an inferior sensitivity and a very slightly inferior specificity. The proportion correctly referred to colonoscopy is lower than for CalDetect, while the proportion incorrectly referred to colonoscopy is slightly higher.

Given the above, when coupled with representations for testing among false negatives and IBS patients not responding to IBS therapy who have not previously been scoped this results in the following test costs, with the other costs from downstream modelling of treatment for induction and maintenance of remission yielding the total estimated costs for the cost utility modelling.

The introductory text to table 50, table 50 and figure 25 have been updated. Please refer to the Diagnostics Assessment Report Addendum

Table 49. Primary Care: Base case results

Comparators	QALYs	Tests	Other	Total
GP no FC				
CD	0.1832	£22	£493	£515
UC	0.2771	£32	£144	£176
IBS	5.7682	£202	£2,404	£2,606
Total	6.2285	£257	£3,041	£3,297
POCT: CalDetect 15µg/g				
CD	0.1832	£23	£493	£516
UC	0.2771	£33	£144	£177
IBS	5.7691	£114	£2,408	£2,522
Total	6.2293	£170	£3,044	£3,214
ELISA				
CD	0.1831	£23	£492	£515
UC	0.2770	£34	£143	£177
IBS	5.7690	£116	£2,407	£2,524
Total	6.2291	£173	£3,042	£3,215

Within the above, in part due to the quite low base case prevalence assumed for IBD within the presenting population, the average QALYs and downstream costs of treatment are broadly in line between the three comparators. There are very slight differences between the comparators' QALYs, with very slight gains from CalDetect over ELISA, and larger though still slight QALY gains over the GP with no faecal calprotectin testing. But the main differences are in the up-front average test costs with CalDetect and ELISA having similar test costs, both of which are somewhat less than those of the GP in the absence of faecal calprotectin testing due to their superior specificity.

The central estimates and cost effectiveness acceptability frontiers (CEAFs) from the probabilistic modelling run over 1,000 iterations are as follows. Within this, it should be borne in mind that the prevalence of IBD is also treated as being probabilistic within the PSA.

The introductory text to table 50, table 50 and figure 25 have been updated. Please refer to the Diagnostics Assessment Report Addendum

Table 50. Primary care: Probabilistic modelling central estimates

	Base case	
	QALYs	Costs
GP	6.1970	£2,028
POCT	6.1978	£1,944
ELISA	6.1975	£1,944

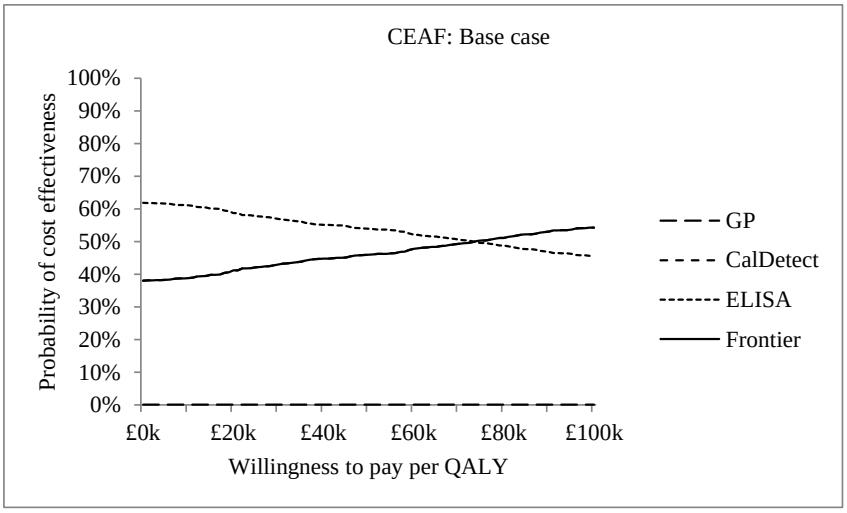


Figure 25. CEAFs: Primary care: Base Case

In the above, the probability of being cost-effective for the comparator of the GP without faecal calprotectin testing never rises above the horizontal axis: i.e. it is estimated that there is no probability of GP without calprotectin testing being cost-effective compared to calprotectin testing.

As for the deterministic modelling, the probabilistic model central estimates suggest small QALY gains from faecal calprotectin testing coupled with cost savings compared to GP referrals in the absence of faecal calprotectin testing. There are very minor differences between CalDetect and ELISA in terms of the central estimates for costs and QALYs. Interestingly, the CEAFs suggest that ELISA is the most likely to be cost effective, though again the differences between the two tests are not marked, but that CalDetect has the highest expected net monetary benefit at all but very low willingness to pay values. Further sensitivity analyses that increase the prevalence of IBD suggest that CalDetect will remain at the frontier for all but very low willingness to pay values, but that as the prevalence of IBD rises the likelihood of CalDetect being the most likely to be cost effective increases; i.e. the cross over point moves towards lower willingness to pay values. But perhaps not too much should be read into this given the similarity of the central estimates, and that a few outlier iterations could have a more marked effect than usual.

Sensitivity analyses: Primary Care: Prevalence of IBD

Varying the prevalence of IBD in the presenting patient population between 5% and 25% results in the following.

Table 51. Sensitivity analyses: Primary Care: Prevalence of IBD

	QALYs			Costs		
IBD prevalence	GP	CalDetect	ELISA	GP	CalDetect	ELISA
5%	6.2135	6.2144	6.2142	£3,190	£3,106	£3,107
10%	6.2706	6.2715	6.2711	£3,599	£3,520	£3,521
15%	6.3277	6.3285	6.3280	£4,008	£3,935	£3,935
20%	6.3848	6.3856	6.3849	£4,416	£4,349	£4,348
25%	6.4419	6.4426	6.4418	£4,825	£4,763	£4,762

As in the base case modelling, the GP with no faecal calprotectin testing is estimated to have the smallest overall QALYs and also to cost more than the other comparators for IBD prevalences up to 25%. At an IBD prevalence of 25% the GP with no faecal calprotectin testing is still estimated to result in QALY losses compared to CalDatect, but in very slight patient gains compared to ELISA due to the less than perfect 93.0% sensitivity of ELISA. The average cost savings from ELISA also fall from £83 at a 5% prevalence to £63 at a 25% prevalence as there are lower cost offsets from avoiding fewer incorrect referrals for colonoscopy. But due to the very limited differences in QALY estimates and the additional average £63 cost, at a prevalence of 25% the cost effectiveness of the GP without faecal calprotectin testing compared to ELISA is estimated to be £378k per QALY.

For CalDetect an increase in the prevalence of IBD increases the net QALY gain over ELISA, this mainly being due to fewer false negatives incorrectly receiving treatment for IBS. This also slightly increases the costs of CalDetect, to the extent that if the prevalence of IBD 20% or more it is no longer cost saving compared to ELISA. At a prevalence of 25% the ICER for CalDetect compared to ELISA is estimated to be £1,697 per QALY, but the differences in terms of the net costs and net QALYs between CalDetect and ELISA remain slight.

Sensitivity analyses: Primary Care: Population tested and GP sensitivity and specificity

As reviewed in the clinical effectiveness section, if faecal calprotectin is made available in primary care the patient group being tested may widen beyond that which GPs previously considered for referral. If patient numbers being tested with faecal calprotectin are double those who would previously have been seriously considered for referral in the absence of faecal calprotectin testing being available, this will affect specificity of the scenario of GP referral in the absence of faecal calprotectin testing. Assuming a doubling of the patient group for faecal calprotectin testing compared

to that previously seriously considered for referral, coupled with a small percentage of additional IBD patients within the additional patient group, could suggest a prevalence of only 3.4% among those being tested with faecal calprotectin, and a sensitivity and specificity for GP referral in the absence of faecal calprotectin testing of 94.1% and 89.7% respectively in this wider patient group. This results in the following.

Table 52. Primary Care: Alternative presenting population sensitivity analyses test results

	GP			CalDetect 15µg/g			ELISA		
	1st test	Colon.	Final	1st test	Colon.	Final	1st test	Colon.	Final
IBD tested	3.4%	3.2%		3.4%	3,4%		3.4%	3.1%	
True +ve	3.2%	3.0%	3.0%	3.4%	3.2%	3.2%	3.1%	3.0%	3.0%
False -ve	0.2%	0.2%	0.4%	0.0%	0.2%	0.2%	0.2%	0.2$	0.4%
IBS tested	96.6%	9.9%		96.6%	5.3%		96.6%	5.8%	
True -ve	86.7%	9.9%	96.6%	91.3%	5.3%	96.6%	90.8%	5.8%	96.6%
False +ve	9.9%	0.0%	0.0%	5.3%	0.0%	0.0%	5.8%	0.0%	0.0%

With the following results from the full cost utility modelling.

Table 53. Primary Care: Alternative presenting population sensitivity analyses test results

Comparators	QALYs	Tests	Other	Total
GP no FC				
CD	0.0973	£12	£261	£273
UC	0.1472	£17	£76	£94
IBS	5.9507	£129	£2,482	£2,612
Total	6.1952	£158	£2,820	£2,978
POCT: CalDetect 15µg/g				
CD	0.0973	£12	£262	£274
UC	0.1473	£18	£76	£94
IBS	5.9510	£118	£2,484	£2,601
Total	6.1956	£147	£2,822	£2,969
ELISA				
CD	0.0973	£12	£261	£273
UC	0.1472	£18	£76	£94
IBS	5.9510	£120	£2,483	£2,604
Total	6.1955	£150	£2,821	£2,971

Faecal calprotectin testing within the wider patient population increases the absolute number of false positives, while by construction it has been assumed to remain constant for those assessed by the GP in the absence of faecal calprotectin testing. This tends to reduce the difference between the costs of the test sequences between calprotectin testing and GP assessment in the absence of faecal calprotectin testing. But despite the assumed doubling in the size of the patient group from those who would seriously be considered for referral by the GP in the absence of faecal calprotectin testing to those who would be tested were faecal calprotectin made available to GPs, the introduction of faecal calprotectin testing is still estimated to be cheaper or at worst broadly cost neutral compared to the previous situation of faecal calprotectin testing not being available. Small QALY gains still accrue from faecal calprotectin testing as well.

Sensitivity analyses: Primary Care: Test cut-offs

For the cut-offs for CalDetect reported within Otten et al the following results are estimated.

Table 54. Sensitivity analysis: Primary Care: Otten et al (2008) CalDetect cut-offs

	QALYs	Costs
15µg/g	6.2293	£3,214
60µg/g	6.2281	£3,187

The slightly better specificity of the 60µg/g cut-off results in slight cost savings of £27 compared to the 15µg/g cut-off, but gains of 0.0012 QALYs are anticipated from the 15µg/g cut-off. This suggests a cost effectiveness of £23,635 per QALY for the 15µg/g cut-off which could be seen as borderline cost effectiveness. Whether the 60µg/g cut-off with an estimated sensitivity of only 61% would be acceptable in practice is a doubtful. Note also the ICER is almost exactly inversely proportionate to the prevalence if IBD in the presenting population; i.e. if it doubles, the ICER halves.

For the cut-offs for CalDetect reported within Hessells et al the following results are estimated.

Table 55. Sensitivity analysis: Primary Care: Hessells et al (2012) CalDetect cut-offs

	QALYs	Costs
15µg/g	6.2269	£3,496
60µg/g	6.2278	£3,352

The results from the estimates of Hessells et al for CalDetect are almost the opposite of those estimated using those of Otten et al. CalDetect with a cut off of 60µg/g is estimated to dominate CalDetect with a cut off of 15µg/g. The specificity is notably better for 60µg/g at 74.2% compared to

53.2% for 15µg/g, and the sensitivity while worse at 87.0% is still somewhat closer to the 95.7% for 15µg/g than the corresponding figures within Otten et al. But it should be borne in mind that Hessells et al did not fit the eligibility criteria of the clinical effectiveness review.

For the cut-offs for Quantum Blue reported within Hessells et al the following results are estimated.

Table 56. Sensitivity analysis: Primary Care: Hessells et al (2012) Quantum Blue cut-offs

	QALYs	Costs
30µg/g	6.2278	£3,390
40µg/g	6.2285	£3,290
50µg/g	6.2283	£3,290
60µg/g	6.2282	£3,267

As would be anticipated the 50µg/g cut-off is dominated, as is the 30µg/g cut-off, by the 40µg/g cut-off. The 40µg/g cut-off is estimated to cost an additional £24 on average compared to the 60µg/g cut-off, while small QALY gains of 0.0003 QALYs suggest an ICER of around £87k. But these QALY differences are extremely minor and the ICER will swing possibly quite wildly if the underlying inputs and assumptions are changed. Note also that the better specificity of the 60µg/g cut-off could have resulted in some further QALY gain from avoidance of the minor adverse effects of colonoscopy had these been included in the modelling. But the better sensitivity of the 40µg/g cut-off would see increases in the prevalence of IBD increase its net QALYs further, the converse being true for a lower IBD prevalence.

For the cut-offs for ELISA reported within Basumani et al the following results are estimated.

Table 57. Sensitivity analysis: Primary Care: Basumani et al (2012) ELISA cut-offs

	QALYs	Costs
50µg/g	[redacted]	[redacted]
100µg/g	[redacted]	[redacted]
150µg/g	[redacted]	[redacted]

The 100µg/g cut-off is only slightly inferior to the 150µg/g cut-off in terms of specificity, but better in terms of sensitivity. This leads to a very slight QALY gain, but an additional £30 cost. The poor specificity of the 50µg/g cut-off leads to it being dominated by the 100µg/g cut-off.

Other sensitivity analyses: Primary Care:

The univariate sensitivity analyses results in the following estimates.

Table 58. Primary care: Univariate sensitivity analyses

	QALYs					Costs				
	GP S1	POCT S2	ELISA S3	net S2 - S1	net S2 - S3	GP S1	POCT S2	ELISA S3	net S2 - S1	net S2 - S3
Base case	6.2285	6.2293	6.2291	0.0009	0.0002	£3,297	£3,214	£3,215	-£83.17	-£1.48
8 week represent	6.2312	6.2320	6.2319	0.0009	0.0002	£3,304	£3,218	£3,220	-£86.21	-£2.08
16 week represent	6.2258	6.2266	6.2263	0.0009	0.0003	£3,274	£3,191	£3,192	-£83.27	-£1.38
24 week represent	6.2204	6.2212	6.2208	0.0009	0.0005	£3,229	£3,146	£3,147	-£83.39	-£0.77
Utilities non-Stark	6.6371	6.6377	6.6376	0.0006	0.0001	£3,297	£3,214	£3,215	-£83.17	-£1.48
No OP	6.2285	6.2293	6.2291	0.0009	0.0002	£3,251	£3,191	£3,192	-£59.88	-£0.73
IBS NR 0%	6.2437	6.2445	6.2443	0.0009	0.0002	£3,281	£3,195	£3,196	-£86.37	-£1.58
IBS NR 10%	6.2133	6.2141	6.2139	0.0008	0.0002	£3,313	£3,233	£3,235	-£79.96	-£1.37
No colon. mort.	6.2286	6.2294	6.2292	0.0008	0.0002	£3,297	£3,214	£3,216	-£83.20	-£1.48

The changes appear to broadly affect the three comparators in a like manner, such that while the estimates of costs and QALYs change there is only a limited impact upon net costs and net QALYs. Faecal calprotectin testing remains cost saving compared to no faecal calprotectin testing, and confers some small additional patient benefits. The costs of the POCT CalDetect faecal calprotectin testing and ELISA faecal calprotectin testing remain very similar throughout, with very slight patient gains from POCT CalDetect faecal calprotectin testing being estimated.

Base case results: Secondary care – IBD versus non-IBD

For the primary care base case, the patient numbers receiving the initial test and being referred for colonoscopy are as below.

Table 59. Secondary Care: Base case results of initial test sequence

	Colonoscopy			ELISA 50µg/g			ELISA 100µg/g		
	1st test	Colon.	Final	1st test	Colon.	Final	1st test	Colon.	Final
IBD tested	..	47.9%		47.9%	47.4%		47.9%	45.0%	
True +ve	..	45.5%	45.5%	47.4%	45.0%	45.0%	45.0%	42.8%	42.8%
False -ve	..	2.4%	2.4%	0.5%	2.4%	2.8%	2.9%	2.3%	5.1%
Non IBD tested	..	52.1%		52.1%	13.5%		52.1%	9.4%	
True -ve	..	52.1%	52.1%	38.6%	13.5%	52.1%	42.7%	9.4%	52.1%
False +ve	..	0.0%	0.0%	13.5%	0.0%	0.0%	9.4%	0.0%	0.0%

ELISA with 50µg/g cut-off results in 13.5% false positives being referred onward for colonoscopy, or 26.0% of the non-IBD patient population as would be anticipated given the 74.0% specificity. This is at the minor cost of 0.5% false negatives not being referred on for colonoscopy at first presentation, or 1.0% of the IBD patient population as would be anticipated given the 99.0% sensitivity. The final results after colonoscopy are 45.0% true positives, 2.8% false negatives and 52.1% true negatives.

ELISA with 100µg/g cut-off results in only 9.4% false positives being referred onward for colonoscopy, or 18.0% of the non-IBD patient population as would be anticipated given the 82.0% specificity. This is at the slightly larger cost of 2.9% false negatives not being referred on for colonoscopy at first presentation, or 6.0% of the IBD patient population as would be anticipated given the 96.0% sensitivity. The final results after colonoscopy are 42.8% true positives, 5.1% false negatives and 52.1% true negatives.

The differences between the two ELISA cut-offs are more marked in terms of false positives with the ELISA cut-off of 50µg/g cut-off resulting in 13.5% false positives being referred to colonoscopy compared to 9.4% for the 100µg/g cut-off. But there is also a difference in the end results in terms of true positives being diagnosed at first presentation: 45.0% for the 50µg/g cut-off and 42.8% for the 100µg/g cut-off, a net difference of 2.3% of the presenting population or 4.4% of the presenting IBD population. Given the above this results in the following estimates

Table 60. Secondary Care: Base case results

Comparators	QALYs	Tests	Other	Total
Colonoscopy				
CD	2.5773	£244	£6,938	£7,183
UC	0.8942	£83	£463	£546
Non-IBD	3.2094	£338	£629	£967
Total	**6.6809**	**£665**	**£8,031**	**£8,696**
ELISA 50µg/g				
CD	2.5767	£254	£6,934	£7,188
UC	0.8941	£86	£463	£549
Non-IBD	3.2117	£120	£634	£754
Total	**6.6824**	**£460**	**£8,031**	**£8,491**
ELISA 100µg/g				
CD	2.5757	£256	£6,921	£7,177
UC	0.8938	£87	£462	£549
Non-IBD	3.2119	£95	£634	£729
Total	**6.6814**	**£438**	**£8,018**	**£8,456**

Faecal calprotectin testing with ELISA is estimated to be both cost saving and more effective than all patients receiving a colonoscopy. There are limited differences between the two ELISA cut-offs, with the 50µg/g cut-off being slightly more expensive on average by £35, due to an additional average £22 for tests among non-IBD patients and an additional average £13 among Crohn's disease patients due to earlier diagnosis. It is also marginally more effective by 0.001 QALYs which suggests a cost effectiveness estimate of £33,982 per QALY, but it should be stressed that the estimated net QALYs are extremely small and that any change in the underlying inputs would have a large swing effect upon the ICER.

The central estimates and CEAFs from the probabilistic modelling run over 1,000 iterations are as follows.

Table 61. Secondary care: Probabilistic modelling central estimates

	Base case	
	QALYs	Costs
Colonoscopy	6.6960	£8,553
ELISA 50µg/g	6.6975	£8,348
ELISA 100µg/g	6.6965	£8,313

The central estimate of net cost of ELISA with the 50µg/g cut-off compared to ELISA with the 100µg/g cut-off remains in line with the deterministic modelling at £35 as are the net QALYs at 0.001. The probabilistic central estimate of the cost effectiveness of ELISA with the 50µg/g cut-off compared to ELISA with the 100µg/g is £33,088 per QALY.

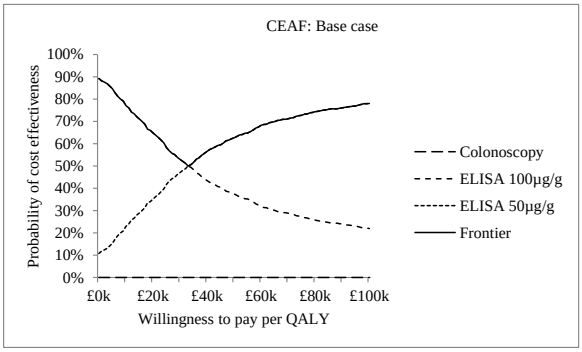


Figure 26. CEAFs: Secondary care: Base Case

Up to a willingness to pay of around £30,000 per QALY it is estimated that the ELISA with the 100µg/g cut-off is most likely to be cost effective and has the highest monetised health benefits net of costs. Thereafter, as the willingness to pay rises further it is estimated that the ELISA with the 50µg/g cut-off is most likely to be cost effective and has the highest monetised health benefits net of costs.

Sensitivity analyses: Secondary Care:

The univariate sensitivity analyses for secondary care result in the following.

Table 62. Secondary care: Univariate sensitivity analyses

	QALYs					Costs				
	Colon. S1	50µg S2	100µg S3	net S2 - S1	net S2 - S3	Colon. S1	50µg S2	100µg S3	net S2 - S1	net S2 - S3
Base case	6.6809	6.6824	6.6814	0.0015	0.0010	£8,696	£8,491	£8,456	-£205	£35.21
40% IBD prev.	6.5950	6.5970	6.5962	0.0020	0.0008	£7,569	£7,330	£7,292	-£239	£37.11
60% IBD prev.	6.8127	6.8135	6.8121	0.0008	0.0014	£10,425	£10,271	£10,239	-£154	£32.28
8 week represent	6.6828	6.6845	6.6839	0.0017	0.0006	£8,707	£8,493	£8,463	-£214	£30.34
16 week represent	6.6789	6.6805	6.6789	0.0015	0.0016	£8,689	£8,479	£8,442	-£209	£37.18
Utilities non-Stark	7.2055	7.2069	7.2066	0.0014	0.0003	£8,696	£8,491	£8,456	-£205	£35.21
No colon. mort.	6.6815	6.6829	6.6818	0.0013	0.0011	£8,697	£8,492	£8,456	-£205	£35.21

As for primary care, most of the changes appear to broadly affect the three comparators in a like manner. The main difference arises from varying the prevalence of IBD, which tends to reduce the cost savings from faecal calprotectin testing as the prevalence rises, as would be anticipated. The source of utilities also has an impact upon the anticipated net gain from ELISA with the 50µg/g cut-off compared to ELISA with the 100µg/g cut-off, the ICER for which worsens to £117k per QALY. But this may be to overstate the effect given the prevalence of Crohn's disease within the presenting population and the perhaps rather small quality of life decrement sourced from Gibson et al.[128]

Summary and discussion

Previous economic analyses have typically concluded that faecal calprotectin testing is cost saving compared to the situation without it. Given test specificities and the assumed prevalences of IBD in the presenting population, the additional cost of the faecal calprotectin testing is more than offset by the reduction in the cost of unnecessary colonoscopies. The YHEC 2010 report for the Centre for Evidence Based Purchasing concluded that faecal calprotectin testing not only saved money through the diagnostic pathway, but that it also resulted in more true positives and true negatives due to its superior sensitivity and specificity and so dominated the situation of the GP referral in the absence of faecal calprotectin testing.

Sensitivities

A distinction of the EAG modelling from that of the literature is that for primary care the GP in the absence of faecal calprotectin testing is estimated to have at least as good a sensitivity as the faecal

calprotectin tests, and for some comparisons a better sensitivity. In this circumstance, the GP results in as many or more true positives than faecal calprotectin testing. As a consequence, despite quite large cost savings and fewer false positives still being estimated for faecal calprotectin testing compared to the GP referring in the absence of faecal calprotectin testing, dominance for faecal calprotectin testing cannot be definitively concluded on the basis the diagnostic pathway alone. There is an argument that the cost and QALY impacts among the false negatives also need to be considered.

The costs and QALY impacts among the false negatives is in the first instance dependent upon the prevalence of IBD in the presenting population and the sensitivities of the tests under consideration. A low IBD prevalence and high test sensitivities mean that there will be few false negatives, while higher IBD prevalences and lower test sensitivities will increase the number of false negatives and so the importance of considering the costs and QALY impacts among them. These latter are dependent upon the average time spent as false negative prior to re-consideration of IBD within a diagnostic pathway. The QALY impacts are also dependent upon the source of the quality of life estimates for false negative being incorrectly treated and for those correctly diagnosed, the latter requiring quality of life estimates for remission and no remission. In the absence of other data it has been assumed that the quality of life among those remaining as a false negative and being incorrectly treated is the same as among those correctly diagnosed but not in remission. The longer the period of time spent as a false negative and the larger the quality of life gain from achieving remission, the larger the impact of false negatives and so the greater the importance of tests' sensitivities.

Modelling induction of remission and maintenance of remission is eased for the current assessment by the relevant models for both Crohn's disease and ulcerative colitis being available as appendices to the respective clinical guidelines, though the ulcerative colitis guidelines is still under consultation. A key assumption within these is that there is no disease progression, such as the development of fistula, when patients are not in remission. Were this to apply, it would also increase the importance of tests' sensitivities.

Adverse events

The modelling also needs to consider the adverse impacts of unnecessary colonoscopies. Due to data constraints, the cost impacts have been limited to modelling the cost impacts of the relatively rare serious adverse events of bleeds and perforations. The quality of life impacts are limited to the mortality associated with perforations. While perforations are rare, so resulting in a very low mortality rate, the QALY impact of this persists for the duration of the model.

There is evidence from the literature that colonoscopies result in minor adverse events among a reasonable proportion of patients; e.g. de Jonge et al[137] suggest that perhaps around 40% of those

investigated with colonoscopy have some effects persisting 30 days subsequent to the colonoscopy. In common with the CG118 guideline on screening for colorectal cancer with colonoscopy[143], these minor adverse events have not been taken into account in the modelling principally due to a lack of quality of life data. The effects of minor and transient colonoscopy side effects seem unlikely to affect the conclusions for the comparisons of no faecal calprotectin testing with faecal calprotectin testing, but they may take on a greater significance in the context of comparing different faecal calprotectin tests or different cut-offs. Depending upon the prevalence of IBD in the presenting population, inclusion of these minor adverse events would increase the importance of tests' sensitivities.

Primary care modelling

For the primary care base case for diagnosis of IBD versus IBS in an adult population, GP referral in the absence of faecal calprotectin testing is compared with CalDetect at the 15µg/g cut-off and with ELISA at the 50µg/g cut-off. The choice of CalDetect at the 15µg/g cut-off may initially seem surprising, but the data from Otten et al suggests a very much worse sensitivity for the 60µg/g cut-off of only 61% which renders it of questionable clinical relevance. A 6.3% prevalence of IBD is drawn from the Durham primary care data.

Within the total patient population both the GP without faecal calprotectin and the initial Caldetect test identifies all 6.3% of patients with IBD as true +ves. The colonoscopy subsequent to this identifies 6.0% of the 6.3% referred as true +ves, due to its 95% sensitivity. The ELISA test is slightly worse, identifying only 5.9% as true +ves with 0.4% being wrongly classified as false –ves. Of the 5.9% referred to colonoscopy, 5.6% are identified as true +ve resulting in a total of 0.7% false +ves.

Within the total patient population, the GP without faecal calprotectin testing incorrectly identified 19.8% as false +ves requiring referral to colonoscopy. The rates of false +ves incorrectly referred to colonoscopy for CalDetect and ELISA are much lower, 5.1% and 5.6% respectively.

Despite its additional initial test costs, faecal calprotectin testing is estimated to result in cost savings compared to the GP without faecal calprotectin testing: £83 for CalDetect and £82 for ELISA. This is on average per patient. This is due mainly to the lower number of colonoscopies. Small QALY gains of around 0.001 QALYs also accrue, though these are limited since the low prevalence of IBD and the similar high sensitivities of the tests result in relatively few false negatives. Some of the QALY differences accrue from the very slightly lower mortality associated with the lower number of colonoscopies. CalDetect and ELISA are estimated to be broadly equivalent with only minor differences between them. Probabilistic modelling results in similar estimates.

Sensitivity analyses around the base case suggest that faecal calprotectin testing results in patient gains and remains cost saving compared to the GP without faecal calprotectin testing up to an IBD prevalence of 25%. At this point, due to ELISA having a less than perfect sensitivity ELISA starts to result in very slight QALY losses compared to the GP without faecal calprotectin testing, though retains cost savings of around £63 per patient on average. The resulting estimate for the cost effectiveness of the GP without faecal calprotectin testing compared to ELISA is £378k per QALY. Due to its perfect sensitivity, CalDetect remains both more effective and cheaper than the GP without faecal calprotectin testing.

The primary care patient group in whom faecal calprotectin is used may be wider than the data set used for the estimation of the sensitivity and specificity of the GP without faecal calprotectin testing. Doubling the size of this patient group and allowing for some additional IBD patients within the wider patient group results in a lower IBD prevalence of only 3.3%, and also sensitivity and specificity estimates for the GP without faecal calprotectin testing in this wider patient group of 94.1% and 89.7% respectively. Despite this improvement in specificity the GP without faecal calprotectin testing is still estimated to result in higher costs and lower QALYs than both CalDetect and ELISA faecal calprotectin testing, though the margin between the with faecal calprotectin testing and without faecal calprotectin testing is narrows quite significantly.

Other univariate sensitivity analyses suggest that the primary care base case results are reasonably robust. The main sensitivity of the results of CalDetect compared to ELISA arise from changing the source of utilities and shortening the time spent as false negatives. These both tend to reduce the importance of false negatives and so reduce the importance of tests' relative sensitivities, and so reduce the estimated net QALY gain from CalDetect over ELISA. But in all of this, it should be stressed that the QALY differences between the faecal calprotectin tests are very small.

Secondary care

For the secondary care paediatric population for the diagnosis of IBD versus non-IBD, direct referral to colonoscopy is compared with ELISA with the 50µg/g cut-off and ELISA with the 100µg/g cut-off. The base case prevalence of IBD of 47.9% increases the importance of test sensitivities compared to the primary care setting, and so the effect of false negatives upon the modelling outputs. Within the total patient population ELISA with the 50µg/g cut-off refers 47.4% as true +ves for colonoscopy, while ELISA with the 100µg/g cut-off refers 45.0% as true +ves for colonoscopy. Colonoscopy is assumed to have a sensitivity of 95%, so the end diagnosis if all are referred immediately to colonoscopy is 45.5% being diagnosed with IBD. For those referred to colonoscopy by ELISA with the 50µg/g cut-off 45.0% are diagnosed as having IBD, while for those referred to colonoscopy by

ELISA with the 100µg/g cut-off 42.8% are diagnosed as having IBD; a net difference between the cut-offs of 2.2%.

Despite the higher IBD prevalence in the secondary care population, the main test differences still lie in the number of unnecessary colonoscopies. Without faecal calprotectin testing all 52.1% of non-IBD patients receive a colonoscopy, compared to 13.5% for the ELISA with the 50µg/g cut-off and only 9.4% for ELISA with the 100µg/g cut-off.

The additional ELISA test costs are more than offset by the savings from reduced colonoscopies. Compared to referring all directly to colonoscopy, ELISA with the 50µg/g cut-off is estimated to save £205 per patient on average, while ELISA with the 100µg/g cut-off is estimated to save £240. Small QALY gains of around 0.001 QALYs are modelling for ELISA compared to direct referral to colonoscopy, these being slightly larger for ELISA with the 50µg/g cut-off due to its better sensitivity. But given the additional average £35 cost, the cost effectiveness estimate for ELISA with the 50µg/g cut-off compared to ELISA with the 100µg/g cut-off is £35,000 per QALY. As before for the primary care modelling, it should be stressed that the QALY differences between the faecal calprotectin tests are very small and perhaps not too much should be read into these differences. The central estimates from the probabilistic modelling are in line with those of the deterministic modelling.

Sensitivity analyses suggest that the base case results are reasonably robust, though the anticipated QALY gain from ELISA with the 50µg/g cut-off compared to ELISA with the 50µg/g cut-off shows some sensitivity the prevalence of IBD, the source of the utilities and the assumed average period of time spent as false negatives as would be expected.

For the modelling in secondary care, compared to the primary care modelling there is additional uncertainty in terms of the model structure. The model is principally a model of IBD versus IBS in an adult population. It may not be as suited to the secondary care paediatric population where the distinction is between IBD and non-IBD. The non-IBD paediatric patients also have a higher proportion of conditions other than IBS compared the adult patient population. But the main differences in terms of costs arise from the up-front test costs, and these will apply within any model construct. A distinction also needs to be drawn between the additional costs of incorrect treatment among false negatives, which for a given set of inputs will be correctly estimated by the model structure, and the structural uncertainty around the appropriate model inputs for Crohn’s disease and for ulcerative colitis in a paediatric population.

Quality of life summary

Table 63. IBS HRQoL studies reporting utilities

Paper	Brazier		Puhan			Spiegel								Wang	
Year	2004		2007			2009								2012	
Country	UK		Canada			USA								Singapore	
Setting	Primary		Gastronterology			General								Symposium	
n	161		96			257	41	82	134	..	..	..	..	198	251
Mean age	47		40			43	n.a.	n.a.	n.a.	..	..	..	..	52	57
Female	86%		84%			79%	n.a.	n.a.	n.a.	..	..	..	..	47%	58%
Mean duration	n.a.		2.7			11.2	n.a.	n.a.	n.a.	..	..	..	..	n.a.	..
Condition	IBS		IBS			All	IBS-C	IBS-D	IBS-M	..	..	..	..	IBS	No IBS
Severity	n.a.		n.a.			Mild-Severe				Non-severe	Severe	3mth relief	3mth no relief	n.a.	..
Measurement	EQ-5D	SF-6D	TTO	SG	SF-6D	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D*	EQ-5D	EQ-5D
HRQoL	0.662	0.666	0.760	0.840	0.850	0.744	0.760	0.760	0.730	0.800	0.700	0.780	0.730	0.739	0.849

*Mapping function unclear

Table 64. IBD HRQoL studies reporting utilities

Paper	Konig						Stark					
Year	2002						2010					
Country	German						German					
Setting	Outpatient	Inpatient					General					
n	121	31					724			723		
Mean age			41	46			41			44		
Female			53%	28%			63%			55%		
Mean duration			15	10			14			13		
Condition			CD	UC			CD			UC		
Severity	79% remiss	7% remiss.	60% remiss.	70% remiss.	Remission	Active	All	Remission	Active	All	Remission	Active
Measurement	EQ-5D^	EQ-5D^	EQ-5D^	EQ-5D^	EQ-5D^	EQ-5D^	EQ-5D	EQ-5D	EQ-5D	EQ-5D	EQ-5D	EQ-5D
HRQoL	0.803	0.619			0.875	0.627	0.770	0.890	0.610	0.840	0.910	0.710

^German mapping function

Paper	Casellas¯	
Year	2005	
Country	Spain	
Setting	OP & IP	
n	628	528
Mean age	34	38
Female	56%	50%
Mean duration	5.0	4.0
Condition	CD	UC

Severity	Remission	Mild	Mod-Severe	Remission	Mild	Mod-Severe
Measurement	EQ-5D¶	EQ-5D¶	EQ-5D¶	EQ-5D¶	EQ-5D¶	EQ-5D¶
HRQoL	0.800	0.720	0.600	1.000	0.720	0.500

¯Median values reported

¶Spanish mapping function

Table 65. Crohn's disease HRQoL studies reporting utilities

Paper	Buxton		Casellas¯
Year	2007		2007
Country	UK		Spain
Setting	Trial		OP
n	n.a.		49
Mean age	n.a.		40
Female	n.a.		47%
Mean duration	n.a.		8.0
Condition	CD		CD
Severity	Mod-Severe		Remission
Measurement	ED-5D	SF-6D	EQ-5D¶
HRQoL	0.700	0.680	1.000

¯Median values reported

¶Spanish mapping function

Paper	Gregor
Year	1997

Country	Canada															
Setting	OP&IP															
n	180		52		34		45		49							
Mean age	35		35		31		34		37							
Female	56%		62%		53%		49%		59%							
Mean duration	8.9		10.3		8.0		7.7		9.3							
Condition	CD		CD		CD		CD		CD		CD		CD		CD	
Severity	All		Chronic refractive		Chronic responsive		Active		Remission		Mild		Mod		Severe	
Measurement	TTO	SG	TTO	SG	TTO	SG	TTO	SG	TTO	SG	TTO	SG	TTO	SG	TTO	SG
HRQoL	0.920	0.810	0.880	0.740	0.980	0.860	0.890	0.770	0.960	0.880	0.955	0.815	0.880	0.725	0.720	0.519

Table 66. Ulcerative colitis HRQoL studies reporting utilities

Paper	Connolly	Bryan			Arsenau			
Year	2009	2008			2006			
Country	W.Europe	UK						
Setting	Trial	HoDAR[J]						
n	127	171						
Mean age	42-47	n.a.						
Female	40%	n.a.						
Mean duration	n.a.	n.a.						
Condition	UC	UC						
Severity	Mild to Mod	Remission	Active	Surgical remiss.	Remission	Active	Surgical remiss.	Surgery comp.
Measurement	EQ-5D	EQ-5D	EQ-5D	EQ-5D	TTO	TTO	TTO	TTO
HRQoL	0.771[I]	0.880	0.420	0.600	0.790	0.320	0.630	0.490

[I] Baseline pooled between arms

[J]Possibly a manufacturer commissioned QoL study

Paper	Poole		
Year	2009		
Country	UK		
Setting	..		
n	126		
Mean age	43		
Female	41%		
Mean duration	..		
Condition	UC		
Severity	Remission	Mild/Mod	Severe
Measurement	EQ-5D	EQ-5D	EQ-5D
HRQoL	0.944	0.811	0.700

4. Discussion

Principal findings

The key findings of this review are;

- In adults, faecal calprotectin is a good indicator of inflammation in the bowel and can be used to distinguish between IBS and IBD in cases where the differential diagnosis is in doubt.
- Calprotectin could be very useful for GPs as a way of confirming a clinical diagnosis of IBS, though it will not be required in all people with IBS, because in some, other features such as a long history, co-morbidities, relationship to stress and an absence of weight loss, may tilt the balance of probability to IBS.
- It is not a perfect test because some patients with IBS have raised calprotectin levels, but false negative IBD is unusual if we use the cut-off of 50 µg/g (for ELISA tests) and 15 µg/g (for Prevent ID POCT) recommended by the manufacturers
- In children, it is useful for distinguishing between IBD and non-inflammatory conditions.
- The balance of risk between sensitivity (not missing any cases of IBD) and specificity (avoiding false positives - people with IBS thought to have IBD) may best be towards sensitivity because missed IBD can lead to much more serious consequences than an unnecessary colonoscopy.
- There are a few patients who have slightly raised levels (50 µg/g to 150 µg, or perhaps to 200ug in children) who may only need monitoring. In many cases, calprotectin level will fall and no further investigation will be necessary. In those who have low-grade IBD, calprotectin will usually rise.
- There are few head to head comparisons of different tests, but such data as there are, do not suggest significant differences in clinical reliability.
- There are no published studies in patients drawn only from primary care
- If calprotectin testing is made available in primary care, GPs will be able to be much more selective in whom they refer to specialist care. Referrals will fall considerably.
- In secondary care, both paediatric and adult, the availability of calprotectin testing will lead to a reduction in the number of colonoscopies performed.
- It is likely that delays in diagnosing IBD will be reduced since a raised calprotectin will alert clinicians. This may be particularly useful in children where the onset can be insidious, as it can also be in some adults.
- Calprotectin testing would lead to cost savings, mainly in secondary care from a reduction in colonoscopies

- Measurement of ESR and CRP in patients with ?IBS, ? IBD, should cease.

Uncertainties

Evidence from primary care

As noted by several commentators, nearly all of the evidence on calprotectin comes from studies from large GI clinics and referral centres.[28,46] The value of the test in primary care is for ruling out IBD, and confirming a presumptive diagnosis of IBS. High sensitivity is therefore required. In theory, sensitivity and specificity are not influenced by prevalence, but they may be if the spectrum of disease alters. However, NPV will be affected, and for GPs is more useful than Se and Sp – a very high NPV will be used to rule out IBD.

Borderline FC results

This usually refers to patients with FC levels in the 50-150 or 200 µg/g range. Most may to come to little harm, may have little visible pathology on endoscopy or video capsule imaging, but some may have very mild CD. They could be monitored for abdominal pain, diarrhoeas and weight loss. The calprotectin test should be repeated at intervals as deemed necessary. The trend over time may be a useful guide to management.

Moroni et al (abstract only) from Glasgow followed up 158 patients newly referred to a GI clinic, aged 16 to 50 with FC values of 50 to 100 µg/g, using the Buhlmann ELISA test.[149] They excluded anyone with known IBD, and anyone who had had a previous FC >100 µg/g. Colonoscopy was carried out in 82, and no IBD was found. There were 6 patients with abnormalities: three with single small (< 10 mm) adenomas, on helminth infection, one diverticulosis and one acute inflammation. They also studies a group with FC < 50 µg/g, and found no IBDs. They conclude that a FC level of <100 µg/g excludes significant pathology.

Zayyat et al (abstract only) from the Kings College group (Gut 2011) linked data from a very large database of FC results, with electronic patient records, to find out what happened to people who had had a borderline FC ("50-100 mg/f", which presumably means 50-100 µg/g), and who had had further investigation with lower GI endoscopy or MRI or CT. Of 433 patients, only 10 had IBD confirmed, and in almost all of these, a repeat FC had shown an increase.[150] The remaining 423 patients were followed up for an average of 3.6 years, but none developed IBD. This suggests that the threshold for action might be raised to 100.

Mohammed and Smale from Leeds (abstract only) report a group of patients who had an abnormal calprotectin (over 50 µg/g), but in whom follow-up endoscopic or radiological investigations were normal.[151] After 3 years, none of those whose baseline calprotectin had been <225 µg/g, developed organic disease. However details in the abstract are limited, with no duration of symptoms prior to first calprotectin testing. Some may have had post-infectious, self-limiting inflammation.

Lee et al from County Durham examined using the manufacturer's recommended cut-off of >60 µg/g in a series of 122 patients. The NPV was high (94%) with only 4 of 71 patients with negative FC being found to have any pathology (though not all were endoscoped), and none of them had IBD. However the results amongst those with positive FC tests were more mixed. 19 of the 51 had organic disease, including nine (18%) with IBD. But 32 (63%) were diagnosed as having functional bowel disorders. All of those with IBD had FC levels well above 100 µg/g (Anjan Dhar, personal communication. 11th March 2013).

Henderson et al from Edinburgh report that in their paediatric group, they tend to regard a cut-off of 200 µg/g as most useful.[26]

Demir et al from King's College Hospital (abstract only) followed up 66 patients with borderline calprotectin (50-150 µg/g) for two years.[152] None developed IBD and calprotectin tended to fall.

Koulaouzidis et al from Edinburgh reported results in a highly selected group of 70 patients suspected of having IBD but in whom both colonoscopy and gastroscopy had found no lesions, and in whom localised small bowel CD was suspected.[153] No patient with calprotectin under 100 µg/g had evidence of CD on small bowel capsule endoscopy, whereas 43% (15/35) of those with calprotectin over 100µg/g were found to have CD, mean 326 µg/g range 116 to 1430 µg/g.

One issue concerning cut-offs is what the sensitivity cut-off should be based on. Should it be based purely on presence of disease – IBD or no IBD? Or should it be based on likely need for treatment? If the latter, perhaps we need two cut-offs and three groups: negative, presumed not to have IBD; positive, IBD requiring treatment; and intermediate, IBD that requires only monitoring meantime.

Manufacturers' current recommended cut-offs appear to be based on the first of the two cut-offs above, and on ensuring that nothing is missed. This seems reasonable in the present state of knowledge.

One crucial issue for the economics is whether someone with mild, or no, symptoms and lowish FC could develop a serious complication such as a stricture or fistula, or develop an ileal mass requiring surgery. Low grade inflammation can continue with little in the way of symptoms. 70-80% of CD patients will require surgery within 5 years of diagnosis. Note that the average time from onset of symptoms to diagnosis of IBD is 12 to 18 months, perhaps longer if symptoms are mild.

One problem for this group if FC was not available would be that GPs might monitor with CRP, ESR and haemoglobin which would all be likely to be normal.

Clinical activity scores

The value of clinical activity scores may be over-estimated. This applies more to monitoring of disease activity after diagnosis. Schoepfer et al surveyed Swiss gastroenterologists and found that most considered clinical activity to be adequate for monitoring disease activity, rather than using markers such as calprotectin.[22] Only 28% of gastroenterologists used calprotectin in more than 70% of their IBD patients. Schoepfer et al concluded that clinical practice in Switzerland was not keeping up with clinical science.

It has been suggested that IBS and IBD may co-exist. This may have arisen because some patients who appear to be in remission according to CDAI and UCAI, have symptoms suggestive of IBS and meet the Rome II criteria. Farrokhyar et al reported that the majority of their patients with "inactive" IBD had symptoms matching the Rome II criteria. The inactivity was based on them not having had a change in therapy for 12 months.[154]

However, the advent of calprotectin has shown that what many of these patients have is on-going inflammation. Keohane et al from Cork studied a group of patients with CD and UC who were apparently in remission, as judged by physicians assessment, CRP <10 mg/l, no treatment in last 6 months, and CDAI 150 or less, or UCAI 3 or less.[155] 60% of the CD group and 39% of the UC group had symptoms that met the Rome II criteria. Calprotectin testing revealed raised levels, indicating that the IBS-like symptoms were due to active IBD. A control group of people with true IBS had normal FC levels. (Note that figure 3 of the paper is wrongly labelled, with IBD that should be IBS.)

In many patients with IBD, symptoms persist. A Finnish study reported that 77% of people with IBD who responded to a survey questionnaire (response rate 40%) had symptoms that impaired quality of life.[156] The mean HRQoL was 169 using the Inflammatory Bowel Disease Questionnaire, which has a range of 32 to 224, with high being better.

Implications of wider use of calprotectin.

There are implications of calprotectin testing for the NICE guidance on drug treatment of CD. TA 187[12] states;

"Infliximab and adalimumab …. are recommended as treatment options for adults with severe Crohn's disease whose disease has not responded to conventional therapy (including immunosuppression and/or corticosteroid treatments)"

Severe is defined by reference to CDAI of 300 or more or Harvey-Bradshaw score of 8 to 9 or more. Paragraph 2.9 notes that;

"The CDAI is frequently used to assess disease severity."

The trials used to underpin the NICE guidance used CDAI as main outcome.

There is no mention of calprotectin in TA 187, because at the time it was written, there was insufficient evidence to support its use. There is an important implication of the use of calprotectin for the NICE guidance. We have noted that clinical scores such as CDAI do not correlate well with mucosal inflammation. We have also noted that some people with CD in apparent remission with "IBS symptoms", have been shown by calprotectin testing to have ongoing inflammation.[157]

Calprotectin testing will reveal a group with few or no symptoms, but on-going inflammation, in whom the anti-TNFs are not recommended. So the present NICE guidance may leave many people with inadequately controlled CD. Treatment of this group may be cost-effective.[24] TA 187 may need to be reviewed in the light of calprotectin data.

Calprotectin is also useful in children, as a non-invasive guide to mucosal inflammation and disease activity in previously diagnosed IBD.[158,159]. Van Rheenen) in an admittedly small series of teenagers with IBD, examined clinical activity indices (PUCAI and PCDAI) CRP and FC for predicting relapse.[160] Calprotectin was more useful than clinical scores, but CRP was not helpful.

The use of calprotectin for monitoring disease activity is outwith the scope of this review and appraisal, but we recommend that the NICE Technology Appraisal Programme should consider when best to assess the impact of calprotectin testing on current guidance on treatment.

Earlier diagnosis and earlier treatment

Would earlier treatment based on a sensitive test to identify inflammation, enable treatment to be started earlier? Could this, by inducing "deep remission" reduce the risk of later complications? (For review, see Panaccione et al.[161] D'Haens et al have reported a close correlation between calprotectin and lesions seen on endoscopy.[162]

As noted above, the current NICE guidance does not recommend biologic agents such as infliximab (which might be regarded as "disease-modifying") until after treatment aimed at relieving symptoms has failed.

Another issue is that people with IBS may have a succession of different treatments, possibly involving several therapeutic trials, as outlined in the Introduction, based on the NICE IBS guidelines. So it might take 6 months or longer before someone with IBD treated as IBS, was recognised as not IBS.

Inflammatory IBS?

Some patients in some studies that were diagnosed as having IBS, after investigation, had raised FC, and it does appear that some people with IBS have an inflammatory component. It has been suggested that this may be due to disturbances in the intestinal bacteria, followed by a mucosal response.[155]

In the County Durham study the range of calprotectin results in those who tested positive (>60 µg/g) but whose final diagnosis was functional bowel disease, was 61 to 547 µg/g (mean 153). [163]

D'Haens et al reported an overlap of calprotectin results between patients with IBD, and a group with IBS: range for IBS 16-139 µg/g.[162]

Therefore, some people with IBS are positive on calprotectin testing. Are they "false positives" or does IBS represent a mix of conditions, some of which have inflammation?

One possibility could be if people with IBS use NSAIDs (including over the counter ibuprofen) that raise calprotectin. However this cannot be the sole explanation, because raised calprotectin levels have been reported in studies that exclude people using NSAIDs.[55,82]

As noted in a previous assessment report for NICE, some people with IBS-D may have bile acid malabsorption.[164] This is a condition in which bile acids are not absorbed as they usually mostly (90%) are in the ileum. The SeHCAT report has no mention of calprotectin. It does not appear to be raised in BAM (except in those whose BAM is due to Crohn's disease in the bile acid absorption site in the ileum) and so that will not be a source of false positives.

Others with IBS may have it subsequent to infectious gastroenteritis, where calprotectin would be raised during the infectious episode, but would then be expected to return to normal. Or does inflammation sometimes continue?

The answer is this group may be repeat testing. In those with raised calprotectin after bowel infection, the level will fall after a few months. However there does appear to be a small number of patients (under 1%) with no evidence of IBD after thorough investigation (Anjan Dhar, personal communication).

There is also a small group with IBS and mild inflammation that responds to NSAIDs (Nick Read, personal communication).

The use of calprotectin in routine care

Trials and other studies may be prone to patient selection bias, and may be an imperfect guide to the use of calprotectin testing in routine care. As previously mentioned, we have data from two pilots of implementation. As of 24th March, we are unable to use most of the data because it has been classed as confidential by NTAC. However data being presented at the British Society for Gastroenterology (Dhar et al, personal communication) show that considerable savings can be made. The Cardiff data show that a considerable proportion of referrals by GPs are to confirm IBS (by exclusion of IBD).[32] So the main value of calprotectin testing may be to confirm presumptive diagnoses of IBS, and that can be done in General Practice. Other studies (Rotherham unpublished) report that over 60% of colonoscopies in the "pre-calprotectin era" showed no pathology.

Alrubaiy et al from Llanelli report results in 74 patients referred to a DGH with intestinal symptoms.[165] Depending on local practice, some had colonoscopy with, or before, calprotectin testing. Two were confirmed to have IBD and both had raised calprotectin levels (mean 271 µg/g). Another 14 had raised levels but further investigations were normal. In the group of 18 that had colonoscopy before calprotectin testing, all colonoscopies were normal, but calprotectin was tested later because of continuing symptoms. In six of the 18, calprotectin was raised, but not by much – mean was 114µg/g. All were finally diagnosed as having no IBD. In the group of 23 that had calprotectin measured before colonoscopy, it was raised in 8 (mean 171 µg/g) who did not have confirmed IBD.

Taylor et al from the Isle of Wight present some data from a small audit. 23 patients had calprotectin >50µg/g, of whom 18 had endoscopy.[166] Only 6 of these had IBD, and all had calprotectin levels >200, with all but one having levels over 300µg/g. However one patient with an initially negative calprotectin later developed IBD.

A key implication of calprotectin testing is the likely reduction in the number of colonoscopies required. Without it, many patients with IBS will be endoscoped. Data from various centres show that over 60% of colonoscopies under the age of 45 show no abnormal findings. Data from Newark and Sherwood show 64% of colonoscopies were normal. (Newark and Sherwood Clinical Commissioning Group 2012 – personal communication)

Results of studies reported earlier, and the consensus of the NICE expert panel, suggest that a positive POCT test is sufficient as a guide to referral, without quantitative ELISA testing. The latter may be done as a baseline for assessing need for, or response to, treatment.

Other tests

S100A12

The S100A12 protein is part of the S100 superfamily also known as calgranulin C. Unlike calprotectin (part of the S100A8/9 family), it is derived exclusively from granulocytes and and monocytes.[167] This has resulted in the suggestion it is perhaps more specific than calprotectin in distinguishing inflammatory related conditions compared with functional types e.g. irritable bowel syndrome (IBS).[82] However both markers show correlation with endoscopic and histological inflammation.[168,169]

The reported ranges for sensitivity with S100A12 are (86 - 97%) and specificity (92-100%).[82,168,169] Cut off most commonly used was 10 μg/g. However it should be noted that unlike calprotectin, most of the studies performed on S100A12 were in children and on a much smaller dataset.

Calprotectin levels are consistently raised in children and fall to reach adult levels by the age of five but there is a suggestion rather that S100A12 does not correlate with age which may suggest an advantage in certain age groups.[82] These calgranulin peptides are also raised in pseudo-inflammatory conditions example colorectal cancer, colorectal polyps and during use of non-steroidal anti inflammatory drugs. Such findings have been reported with calprotectin [170,171], but little is known of its effects on expression of S100A12.

Thus if S100A12 were to be considered an alternative marker to distinguish between inflammatory bowel disease and IBS, further larger studies would need to be performed (especially in adults) and to determine its expression in other gut related conditions. One particular area would be to confirm

reports that unlike calprotectin, S100A12 is elevated in bacterial gastroenteritis and not viral gastroenteritis which may prove useful in clinical practice.[168]

Faecal haemoglobin

Mooiweer et al suggested that, at least in surveillance of IBD, faecal haemoglobin might be an alternative to calprotectin. Their study was based on findings in 119 patients having surveillance colonoscopy.[172] Faecal haemoglobin was as accurate an indicator of inflammation as calprotectin. However the mean calprotectin level in patients with active inflammation was 451 µg/g, so the spectrum of disease is rather different from the patients in the NICE decision problem group.

Lactoferrin

NICE did not include lactoferrin in the scope of this appraisal. There is less evidence on lactoferrin than on calprotectin. Testing for lactoferrin uses mainly ELISA methods but a point of care test is also available.[65] Lactoferrin is an iron-binding protein present in neutrophils.[173] Faecal lactoferrin (FL) is stable at room temperature for 4 days.[174] There are suggestions that FL is as good as FC for differentiating inflammatory bowel disease from non-inflammatory bowel disease.[174].

For differentiating IBD from IBS, the sensitivity of lactoferrin ranged between 78 and 82% while the specificity ranged between 85 and 100% in studies.[173] Schoepfer et al compared the accuracy of faecal markers, blood leukocytes, CRP and IBD antibodies in discriminating IBD from IBS and found that the overall accuracies of FC ELISA (Phical test – 89%) and lactoferrin (IBD-SCAN – 90%) were similar.[67] In another study[175] using colonoscopy as the reference standard, the sensitivity, specificity, PPV and diagnostic efficacy were slightly higher with lactoferrin (80, 85, 87 and 81%) than with calprotectin (78, 83, 86 and 80%).

Otten et al[65] compared the diagnostic performance of the two new rapid tests for faecal lactoferrin (FL) and FC against ELISA and also assessed their potential to differentiate IBD from IBS. The sensitivity and NPV for the FC rapid test were higher than the FL rapid test (100% vs. 78%, 100% versus 95%), whereas specificity and PPV were higher for the FL rapid test than the FC rapid test (99%, 95% vs. 95%, 82%). The diagnostic accuracy for both rapid tests was similar to ELISA tests (Cohen's kappa - 0.69 for FC; 0.68 for lactoferrin). Schroder et al [69] found that the sensitivity with FC was better than FL (93% versus 82%) while specificity was 100% for both. One study [176] compared the use of FC and FL as noninvasive markers in children and young adult with IBD and found that using both FC and FL as diagnostic tests was better than using them in isolation. In contrast, another study [69] found that using two tests together did not provide additional benefit.

The above findings suggest that FL can be used to differentiate IBD from IBS. The new FL rapid test seems to be as good as the ELISA in differentiating IBD from IBS thus there may be a place for this test in primary care. However, further research in primary care populations should compare the rapid FL test against rapid FC tests.

Neopterin

Nancey et al reported a comparison of neopterin and calprotectin, with endoscopic scores.[177] Both distinguished active from inactive IBD, though with better accuracy in UC than CD. Neopterin was as accurate as calprotectin.

Ongoing research and research needs.

Arasaradnam et al from Coventry and Rotherham have pioneered an "electric nose" test for IBD detection using urine testing to distinguish between those in remission and those not, and between those with IBD and healthy controls.[178] This could potentially be used for diagnosis but requires a comparison of people with newly presenting IBD and those with IBS. The rationale behind this test is that abnormal gut permeability allows fermentation breakdown products into the bloodstream and hence into urine.

A Canadian group is carrying the FOCUS study (The Future of Fecal Calprotectin Utility Study: NCT0167324) to find out how often calprotectin results would change management of patients.[179]

Some patients with IBS (diagnosed after negative endoscopies) have raised calprotectin, The reasons for this are not known, and research into this group may be indicated. It may be due to an inflammatory component in some patients with IBS, perhaps especially those whose IBS follows gastrointestinal infection.

Uncertainty remains as to how to deal with patients who have calprotectin levels between 50 and 200 µg/g. Repeat testing after 6-8 weeks is recommended.

Comparative data on the relative performance of the POCT tests, including in the intermediate 50 to 200 µg/g zone, is required.

Conclusions

Faecal calprotectin testing appears to be a useful method of distinguishing between inflammatory and non-inflammatory chronic bowel disease.

The current evidence base does not suggest any preference for any test over the others on diagnostic grounds. Relative cost will be more important in choice of test. The test kits are not expensive and relative cost may depend more on labour costs and local discounts.

The NICE scope raised questions, abbreviated in italics below.

Is calprotectin testing a reliable way of differentiating inflammatory disease of the bowel from non-inflammatory ones?
Yes. Faecal calprotectin testing identifies patients with inflammation of the bowel, who need referral to specialist care. The majority of younger adult patients seen with lower abdominal symptoms in general practice have IBS, and the absence of inflammation as indicated by a negative calprotectin test means that IBD is very unlikely. They can then be managed in primary care and spared further investigations.

What are the optimal cut-offs for use in primary and secondary care
The same cut-off should be used in primary and secondary care – 50µg/g. This is based on ensuring high sensitivity, and not missing people with IBD. Some people assessed as positive by this cut-off will have borderline levels of 50 to 200µg/g, and may initially be monitored with repeat calprotectin testing, but some of this group will progress to definite IBD.

How do the rapid point-of-care tests compare to the laboratory tests?
There are few studies directly comparing tests, and on clinical effectiveness grounds, there is insufficient evidence to recommend one test over the others. The point of care tests can provide faster results. Costs vary amongst tests. None of the test kits are expensive but labour costs vary. The evidence base varies amongst tests. There are currently no grounds on either diagnostic reliability or cost-effectiveness considerations for preferring one test over another.

How will calprotectin testing perform in primary care?
Sensitivity and specificity will be as good in primary care, but the lower prevalence will increase the negative predictive value. The main benefit in primary care will be to confirm the clinical diagnosis by GPs of IBS. Making calprotectin testing available to general practitioners will greatly reduce the number of younger adults referred to specialist care, and the need for invasive investigations such as colonoscopy.

Impact in secondary care

In secondary care, the main benefit will be a marked reduction in colonoscopies that find no abnormalities. Calprotectin testing will considerably reduce the number of colonoscopies required. In various studies, over 60% of colonoscopies in this group of adult patients have been normal

Calprotectin testing will lead to considerable savings to the NHS, as well as the avoidance of an unpleasant invasive procedure in people whose symptoms are due to IBS.

Calprotectin testing can also reduce the need for colonoscopy in children who do not have IBD, and could reduce diagnostic delays in those who do. It could also reduce loss of work time for parents and loss of school time for children.

References

1. Ford AC, Talley NJ. Irritable bowel syndrome. *BMJ* 2012;345:e5836.

2. Wang YT, Lim HY, Tai D, Krishnamoorthy TL, Tan T, Barbier S *et al*. The impact of Irritable Bowel Syndrome on health-related quality of life: a Singapore perspective. *BMC Gastroenterology* 2012;12:104.

3. Spiegel B, Harris L, Lucak S, Mayer E, Naliboff B, Bolus R *et al*. Developing valid and reliable health utilities in irritable bowel syndrome: results from the IBS PROOF Cohort. *American Journal of Gastroenterology* 2009;104:1984-91.

4. Maxion-Bergemann S, Thielecke F, Abel F, Bergemann R. Costs of irritable bowel syndrome in the UK and US. [Review] [49 refs]. *Pharmacoeconomics* 2006;24:21-37.

5. Akehurst RL, Brazier JE, Mathers N, O'Keefe C, Kaltenthaler E, Morgan A *et al*. Health-related quality of life and cost impact of irritable bowel syndrome in a UK primary care setting. *Pharmacoeconomics* 2002;20:455-62.

6. Badia X, Mearin F, Balboa A, Baro E, Caldwell E, Cucala M *et al*. Burden of illness in irritable bowel syndrome comparing Rome I and Rome II criteria. *Pharmacoeconomics* 2002;20:749-58.

7. British Society of Gastroenterology. Chronic management: IBS/Functional symptoms. 2009 http://www.bsg.org.uk/clinical/commissioning-report/ibs/functional-symptoms.html (accessed 29 March 2013)

8. British Society of Gastroenterology. Clinical Commissioning Report: Diagnosis Chonic Abdominal discomfort, Diarrhoea and Constipation. 2013 http://www.bsg.org.uk/clinical/commissioning-report/chonic-abdominal-discomfort-diarrhoea-and-constipation.html (accessed 6 March 2013)

9. Degen A, Buning C, Siegmund B, Prager M, Maul J, Preiss J et al. Reasons for a delayed diagnosis in inflammatory bowel disease. ECCO 2013 Congress Abstracts 2013 https://www.ecco-ibd.eu/publications/congress-abstract-s.html (accessed 4 February 2013)

10. Ordas I, Eckmann L, Talamini M, Baumgart DC, Sandborn WJ. Ulcerative colitis. *Lancet* 2012;380:1606-19.

11. Baumgart DC, Sandborn WJ. Crohn's disease. *Lancet* 2012;380:1590-605.

12. National Institute for Health and Clinical Excellence. Crohn's disease - infliximab (review) and adalimumab (review of TA40) (TA187). 2010 http://guidance.nice.org.uk/TA187 (accessed 7 March 2013)

13. Henderson P, Hansen R, Cameron FL, Gerasimidis K, Rogers P, Bisset WM *et al*. Rising incidence of pediatric inflammatory bowel disease in Scotland. *Inflamm Bowel Dis* 2012;18:999-1005.

14. Aldhous MC, Armitage E, Drummond H, Satsangi J. Evidence of north-south gradient in incidence of juvenile - Onset inflammatory bowel disease in Scotland. *Gastroenterology* 2003;124:A213.

15. Henderson P, Wilson DC. The rising incidence of paediatric-onset inflammatory bowel disease. *Arch Dis Child* 2012;97:585-6.

16. Shamir R, Phillip M, Levine A. Growth retardation in pediatric Crohn's disease: pathogenesis and interventions. [Review] [80 refs]. *Inflammatory Bowel Dis* 2007;13:620-8.

17. Ford AC, Moayyedi P, Hanauer SB. Ulcerative colitis. *BMJ* 2013;346:f432.

18. Sprakes MB, Ford AC, Suares NC, Warren L, Greer D, Donnellan CF *et al*. Costs of care for Crohn's disease following the introduction of infliximab: a single-centre UK experience. *Alimentary Pharmacology & Therapeutics* 2010;32:1357-63.

19. Waters HC, Vanderpoel JE, Nejadnik B, McKenzie RS, Lunacsek OE, Lennert BJ *et al*. Resource utilization before and during infliximab therapy in patients with inflammatory bowel disease. *Journal of Medical Economics* 2012;15:45-52.

20. Butcher RO, Mehta SJ, Ahmad OF, Boyd CA, Anand R, Stein J *et al*. Incidental diagnosis of inflammatory bowel disease in a British bowel cancer screening cohort: A multi-centre study. *Journal of Crohn's and Colitis* 2013;7:S97-S98.

21. Esch A, Bourrier A, Seksik P, Nion-Larmurier I, Sokol H, Beaugerie L *et al*. What is the prognosis of silent Crohn's disease? *Journal of Crohn's and Colitis* 2013;7:S55.

22. Schoepfer AM, Vavricka S, Zahnd-Straumann N, Straumann A, Beglinger C. Monitoring inflammatory bowel disease activity: clinical activity is judged to be more relevant than endoscopic severity or biomarkers. *J Crohns Colitis* 2012;6:412-8.

23. NZ IBD Research Review. Inflammatory Bowel Disease. 2010 http://www.researchreview.co.nz/index.php?option=com_flexicontent&view=category&cid=82&Itemid=94 (accessed 26 March 2013)

24. Ananthakrishnan AN, Korzenik JR, Hur C. Can Mucosal Healing Be a Cost-effective Endpoint for Biologic Therapy in Crohn's Disease? A Decision Analysis. *Inflamm Bowel Dis* 2013;19:37-44.

25. Shivananda S, Lennard-Jones J, Logan R, Fear N, Price A, Carpenter L *et al*. Incidence of inflammatory bowel disease across Europe: is there a difference between north and south? Results of the European Collaborative Study on Inflammatory Bowel Disease (EC-IBD). *Gut* 1996;39:690-7.

26. Henderson P, Casey A, Lawrence SJ, Kennedy NA, Kingstone K, Rogers P *et al*. The diagnostic accuracy of fecal calprotectin during the investigation of suspected pediatric inflammatory bowel disease. *American Journal of Gastroenterology* 2012;107:941-9.

27. Ford AC, Talley NJ, Veldhuyzen van Zanten SJ, Vakil NB, Simel DL, Moayyedi P. Will the history and physical examination help establish that irritable bowel syndrome is causing this patient's lower gastrointestinal tract symptoms? *JAMA* 2008;300:1793-805.

28. Jellema P, van der Windt DA, Schellevis FG, van der Horst HE. Systematic review: accuracy of symptom-based criteria for diagnosis of irritable bowel syndrome in primary care. *Aliment Pharmacol Ther* 2009;30:695-706.

29. National Institute for Health and Clinical Excellence. Faecal calprotectin diagnostic tests for identifying inflammatory bowel disease: final scope. 2012 http://www.nice.org.uk/nicemedia/live/13789/61043/61043.pdf (accessed 29 March 2013)

30. National Collaborating Centre for Nursing and Supportive Care. Clinical practice guideline: Irritable bowel syndrome in adults: Diagnosis and management of irritable bowel syndrome in primary care. 2008 http://www.nice.org.uk/nicemedia/live/11927/39746/39746.pdf (accessed 29 March 2013)

31. Thompson WG, Heaton KW, Smyth GT, Smyth C. Irritable bowel syndrome: the view from general practice. *Eur J Gastroenterol Hepatol* 1997;9:689-92.

32. Ramadas A, Datta K, Sunderraj L, Hawthorne AB. Faecal calprotectin as part of standardised assessment tool of suspected irritable bowel syndrome (IBS): A novel investigative algorithm [abstract]. *Gut* 2011;60:A73-A74.

33. Kok L, Elias SG, Witteman BJ, Goedhard JG, Muris JW, Moons KG *et al*. Diagnostic accuracy of point-of-care fecal calprotectin and immunochemical occult blood tests for diagnosis of organic bowel disease in primary care: the Cost-Effectiveness of a Decision Rule for Abdominal Complaints in Primary Care (CEDAR) study. *Clin Chem* 2012;58:989-98.

34. Burgmann T, Clara I, Graff L, Walker J, Lix L, Rawsthorne P *et al*. The Manitoba Inflammatory Bowel Disease Cohort Study: prolonged symptoms before diagnosis--how much is irritable bowel syndrome? *Clin Gastroenterol Hepatol* 2006;4:614-20.

35. National Institute for Health and Clinical Excellence. Irritable bowel syndrome (CG61). NICE clinical guideline 61 2008 http://www.nice.org.uk/CG061

36. Naismith GD, Smith LA, Barry SJ, Munro JI, Laird S, Rankin K *et al*. A prospective single-centre evaluation of the intra-individual variability of faecal calprotectin in quiescent Crohn's disease. *Aliment Pharmacol Ther* 2013;37:613-21.

37. Moum B, Jahnsen J, Bernklev T. Fecal calprotectin variability in Crohn's disease. *Inflammatory Bowel Dis* 2010;16:1091-2.

38. Haapamaki J, Tanskanen A, Roine RP, Blom M, Turunen U, Mantyla J *et al*. Medication use among inflammatory bowel disease patients: excessive consumption of antidepressants and analgesics. *Scand J Gastroenterol* 2013;48:42-50.

39. Demir OM, Appleby R, Wang Y, Logan RPH. What factors might contribute to borderline faecal calprotectin levels?[abstract]. *Journal of Crohn's and Colitis* 2012;6:S56.

40. Turvill J. High negative predictive value of a normal faecal calprotectin in patients with symptomatic intestinal disease. *Frontline Gastroenterol* 2012;3:21-8.

41. Whitehead WE, Palsson OS, Feld AD, Levy RL, von KM, Turner MJ *et al*. Utility of red flag symptom exclusions in the diagnosis of irritable bowel syndrome. *Aliment Pharmacol Ther* 2006;24:137-46.

42. Dolwani S, Metzner M, Wassell JJ, Yong A, Hawthorne AB. Diagnostic accuracy of faecal calprotectin estimation in prediction of abnormal small bowel radiology. *Alimentary Pharmacology & Therapeutics* 2004;20:615-21.

43. 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;123:450-60.

44. York Health Economics Consortium. Economic report: Value of calprotectin in screening out irritable bowel syndrome: CEP09041. 2010 http://www.alphalabs.co.uk/asp/db/documents/cep09026_Calprotectin%20Economic_Jan10.pdf (accessed 7 March 2013)

45. Mascialino B, Hermansson L-L, Larsson A. Is the IBD pre-endoscopic screening F-Calprotectin test more cost-effective than the usage of serologic markers in selected European markets? *Journal of Crohn's and Colitis* 2013;7:S91.

46. Burri E, Beglinger C. Faecal calprotectin -- a useful tool in the management of inflammatory bowel disease. [Review]. *Swiss Med Wkly* 2012;142:w13557.

47. van Rheenen PF, Van d, V, Fidler V. Faecal calprotectin for screening of patients with suspected inflammatory bowel disease: diagnostic meta-analysis. *BMJ* 2010;341:c3369.

48. von Roon AC, Karamountzos L, Purkayastha S, Reese GE, Darzi AW, Teare JP *et al*. Diagnostic precision of fecal calprotectin for inflammatory bowel disease and colorectal malignancy. *American Journal of Gastroenterology* 2007;102:803-13.

49. Harbord RM, Whiting P. metandi: Meta-analysis of diagnostic accuracy using hierarchical logistic regression. *Stata Journal* 2009;9:211-29.

50. Macaskill P, Gatsonis C, Deeks J, Harbord RM, Takwoingi Y, (editors). Chapter 10: Analysing and Presenting Results. Cochrane Handbook for Systematic Reviews of Diagnostic Test Accuracy Version 1.0 The Cochrane Collaboration 2010.

51. Harbord R. Meta-analysis of diagnostic test accuracy studies: metandi& midas. 2010 http://www.stata.com/meeting/uk10/UKSUG10.Harbord.pdf (accessed 7 March 2013)

52. Whiting P, Rutjes AW, Reitsma JB, Bossuyt PM, Kleijnen J. The development of QUADAS: a tool for the quality assessment of studies of diagnostic accuracy included in systematic reviews. *BMC Med Res Methodol* 2003;3:25.

53. 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;23:985-92.

54. Garcia SM, V, Gonzalez R, Iglesias FE, Gomez CF, Casais JL, Cerezo RA *et al*. [Diagnostic value of fecal calprotectin in predicting an abnormal colonoscopy]. [Spanish]. *Med Clin* 2006;127:41-6.

55. Li XG, Lu YM, Gu F, Yang XL. [Fecal calprotectin in differential diagnosis of irritable bowel syndrome]. [Chinese]. *Beijing da Xue Xue Bao Yi Xue Ban/Journal of Peking University Health Sciences* 2006;38:310-3.

56. Licata A, Randazzo C, Cappello M, Calvaruso V, Butera G, Florena AM *et al*. Fecal Calprotectin in Clinical Practice A Noninvasive Screening Tool for Patients With Chronic Diarrhea. *J Clin Gastroenterol* 2012;46:504-8.

57. Shitrit AB, Braverman D, Stankiewics H, Shitrit D, Peled N, Paz K. Fecal calprotectin as a predictor of abnormal colonic histology. *Diseases of the Colon & Rectum* 2007;50:2188-93.

58. Zippi M, Al AN, Siliquini F, Severi C, Kagarmanova A, Maffia C *et al*. Correlation between faecal calprotectin and magnetic resonance imaging (MRI) in the evaluation of inflammatory pattern in Crohn's disease. *Clinica Terapeutica* 2010;161:e53-e56.

59. Aomatsu T, Yoden A, Matsumoto K, Kimura E, Inoue K, Andoh A *et al*. Fecal calprotectin is a useful marker for disease activity in pediatric patients with inflammatory bowel disease. *Digestive Diseases & Sciences* 2011;56:2372-7.

60. Canani RB, de Horatio LT, Terrin G, Romano MT, Miele E, Staiano A *et al*. Combined use of noninvasive tests is useful in the initial diagnostic approach to a child with suspected inflammatory bowel disease. *Journal of Pediatric Gastroenterology & Nutrition* 2006;42:9-15.

61. Tomas AB, Vidal MV, Camps R. Fecal calprotectin as a biomarker to distinguish between organic and functional gastrointestinal disease. *Rev Esp Enferm Dig* 2007;99:689-93.

62. York Health Economics Consortium. Evidence review: Value of calprotectin in screening out irritable bowel syndrome: CEP09026. 2010 http://www.alphalabs.co.uk/asp/db/documents/cep09026_Calprotectin%20Evidence_Jan10.pdf (accessed 7 March 2013)

63. Bunn SK, Bisset WM, Main MJ, Gray ES, Olson S, Golden BE. Fecal calprotectin: validation as a noninvasive measure of bowel inflammation in childhood inflammatory bowel disease. *Journal of Pediatric Gastroenterology & Nutrition* 2001;33:14-22.

64. Kolho K, Turner D, Veereman-Wauters G, Sladek M, de RL, Shaoul R *et al*. Rapid test for fecal calprotectin levels in children With Crohn Disease. *J Pediatr Gastroenterol Nutr* 2012;55:436-9.

65. Otten CM, Kok L, Witteman BJ, Baumgarten R, Kampman E, Moons KG *et al*. Diagnostic performance of rapid tests for detection of fecal calprotectin and lactoferrin and their ability to discriminate inflammatory from irritable bowel syndrome.[Erratum appears in Clin Chem Lab Med. 2008;46(12):1798]. *Clinical Chemistry & Laboratory Medicine* 2008;46:1275-80.

66. Schoepfer AM, Trummler M, Seeholzer P, Criblez DH, Seibold F. Accuracy of four fecal assays in the diagnosis of colitis. *Diseases of the Colon & Rectum* 2007;50:1697-706.

67. Schoepfer AM, Trummler M, Seeholzer P, Seibold-Schmid B, Seibold F. Discriminating IBD from IBS: comparison of the test performance of fecal markers, blood leukocytes, CRP, and IBD antibodies. *Inflammatory Bowel Dis* 2008;14:32-9.

68. Tibble JA, Sigthorsson G, Bridger S, Fagerhol MK, Bjarnason I. Surrogate markers of intestinal inflammation are predictive of relapse in patients with inflammatory bowel disease. *Gastroenterology* 2000;119:15-22.

69. Schroder O, Naumann M, Shastri Y, Povse N, Stein J. Prospective evaluation of faecal neutrophil-derived proteins in identifying intestinal inflammation: combination of parameters does not improve diagnostic accuracy of calprotectin. *Alimentary Pharmacology & Therapeutics* 2007;26:1035-42.

70. Limburg PJ, Ahlquist DA, Sandborn WJ, Mahoney DW, Devens ME, Harrington JJ *et al*. Fecal calprotectin levels predict colorectal inflammation among patients with chronic diarrhea referred for colonoscopy. *American Journal of Gastroenterology* 2000;95:2831-7.

71. Jellema P, van Tulder MW, van der Horst HE, Florie J, Mulder CJ, van der Windt DA. Inflammatory bowel disease: a systematic review on the value of diagnostic testing in primary care. [Review]. *Colorectal Dis* 2011;13:239-54.

72. Kostakis ID, Cholidou KG, Vaiopoulos AG, Vlachos IS, Perrea D, Vaos G. Fecal Calprotectin in Pediatric Inflammatory Bowel Disease: A Systematic Review. *Dig Dis Sci* 2012.

73. Perminow G, Brackmann S, Lyckander LG, Franke A, Borthne A, Rydning A *et al*. A characterization in childhood inflammatory bowel disease, a new population-based inception cohort from South-Eastern Norway, 2005-07, showing increased incidence in Crohn's disease. *Scand J Gastroenterol* 2009;44:446-56.

74. Van de Vijver E, Schreuder AB, Cnossen WR, Muller Kobold AC, van Rheenen PF. Safely ruling out inflammatory bowel disease in children and teenagers without referral for endoscopy. *Arch Dis Child* 2012;Sep 27. [Epub ahead of print].

75. Basumani P, Bardhan K, Eyre R, Ellis R, The Rotherham Team. Faecal calprotectin: Rotherham experience [slide presentation]. BSG Away day 28/06/2012

76. El-Badry A, Sedrak H, Rashed L. Faecal calprotectin in differentiating between functional and organic bowel diseases. *Arab J Gastroenterol* 2010;11:70-3.

77. Bharathi S, Moncur P, Holbrook IB, Kelly S. A normal facael calprotectin has a high negative predictive value in cases of suspected irritable bowel syndrome [abstract]. *Gastroenterology* 2005;128:A459.

78. Carroccio A, Iacono G, Cottone M, Di PL, Cartabellotta F, Cavataio F *et al*. Diagnostic accuracy of fecal calprotectin assay in distinguishing organic causes of chronic diarrhea from irritable bowel syndrome: a prospective study in adults and children. *Clin Chem* 2003;49:t-7.

79. Ashorn S, Honkanen T, Kolho KL, Ashorn M, Valineva T, Wei B *et al*. Fecal calprotectin levels and serological responses to microbial antigens among children and adolescents with inflammatory bowel disease. *Inflammatory Bowel Dis* 2009;15:199-205.

80. Diamanti A, Panetta F, Basso MS, Forgione A, Colistro F, Bracci F *et al*. Diagnostic work-up of inflammatory bowel disease in children: the role of calprotectin assay. *Inflammatory Bowel Dis* 2010;16:1926-30.

81. Fagerberg UL, Loof L, Myrdal U, Hansson LO, Finkel Y. Colorectal inflammation is well predicted by fecal calprotectin in children with gastrointestinal symptoms. *Journal of Pediatric Gastroenterology & Nutrition* 2005;40:450-5.

82. Sidler MA, Leach ST, Day AS. Fecal S100A12 and fecal calprotectin as noninvasive markers for inflammatory bowel disease in children. *Inflammatory Bowel Dis* 2008;14:359-66.

83. Shaoul R, Sladek M, Turner D, Paeregaard A, Veres G, Wauters GV *et al*. Limitations of fecal calprotectin at diagnosis in untreated pediatric Crohn's disease. *Inflammatory Bowel Dis* 2012;18:1493-7.

84. Bremner A, Roked S, Robinson R, Phillips I, Beattie M. Faecal calprotectin in children with chronic gastrointestinal symptoms. *Acta Paediatrica* 2005;94:1855-8.

85. Komraus M, Wos H, Wiecek S, Kajor M, Grzybowska-Chlebowczyk U. Usefulness of faecal calprotectin measurement in children with various types of inflammatory bowel disease. *Mediators Inflamm* 2012;2012:608249.

86. Burri E, Manz M, Rothen C, Rossi L, Beglinger C, Lehmann FS. Monoclonal antibody testing for fecal calprotectin is superior to polyclonal testing of fecal calprotectin and lactoferrin to identify organic intestinal disease in patients with abdominal discomfort. *Clin Chim Acta* 2013;416:41-7.

87. Lee H, Mainman H, Borthwick H, Dhar A. Faecal calprotectin testing in primary and secondary care – are the current manufacturer's cut-off levels clinically useful? *unpublished data BSG 2013 abstract submission* 2013.

88. Manz M, Burri E, Rothen C, Tchanguizi N, Niederberger C, Rossi L *et al*. Value of fecal calprotectin in the evaluation of patients with abdominal discomfort: an observational study. *BMC Gastroenterology* 2012;12.

89. Wassell J, Wallage M, Brewer E. Evaluation of the Quantum Blue rapid test for faecal calprotectin. *Ann Clin Biochem* 2012;49:55-8.

90. Dolci A, Panteghini M. Comparative study of a new quantitative rapid test with an established ELISA method for faecal calprotectin. *Clin Chim Acta* 2012;413:350-1.

91. Coorevits L, Baert F, Vanpoucke H. Comparative study of the Quantum Blue rapid test for point of care quantification of faecal calprotectin with an established ELISA method. *Journal of Crohn's and Colitis* 2012;6:S45.

92. Hessels J, Douw G, Yildirim DD, Meerman G, Van Herwaarden MA, Van Den Bergh FAJT. Evaluation of Prevent ID and Quantum Blue rapid tests for fecal calprotectin. *Clin Chem Lab Med* 2012;50:1079-82.

93. 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. *Journal of Crohn's & colitis* 2012;6:207-14.

94. Vestergaard TA, Nielsen SL, Dahlerup JF, Hornung N. Fecal calprotectin: assessment of a rapid test. *Scandinavian Journal of Clinical & Laboratory Investigation* 2008;68:343-7.

95. Shastri Y, Schroder O, Stein J. Comparative evaluation of a new semi-quantitative, rapid, Office-based strip test with an ELISA-based assay for measuring fecal calprotectin to assess intestinal inflammation: Prospective multicenter clinical study. *Gastroenterology* 2009;136:A34-A35.

96. Labaere D, Smismans A, Van OG, Christiaens P, D'Haens G, Moons V *et al*. Clinical comparison of eight different calprotectin immunoassays for the diagnosis and follow-up of inflammatory bowel disease. *Journal of Crohn's and Colitis* 2013;7:S120.

97. 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.

98. Loitsch SM, Berger C, Hartmann F, Stein J. Comparison of two commercially available serologic kits for the detection of fecal calprotecin [abstract]. *Gastroenterology* 2010;138:S528.

99. Tomkins C, Zeino Z, Nwokolo C, Smith SC, Arasaradnam R. Faecal calprotectin analysis: does the method matter? [meeting abstract]. *Gut* 2012;61:A173-A174.

100. Spiller R, Aziz Q, Creed F, Emmanuel A, Houghton L, Hungin P *et al*. Guidelines on the irritable bowel syndrome: mechanisms and practical management. *Gut* 2007;56:1770-98.

101. Curtis L. Personal Social Services Research Unit (PSSRU): Unit Costs of Health and Social Care. 2011 http://www.pssru.ac.uk/project-pages/unit-costs/2011/index.php (accessed 7 March 2013)

102. National Institute for Health and Clinical Excellence. Crohn's disease (CG152). NICE clinical guideline 152 2012 guidance.nice.org.uk/cg152 (accessed 7 March 2013)

103. National Institute for Health and Clinical Excellence. Ulcerative colitis: management in adults, children and young people: NICE guideline.Draft for consultation. 2013 http://guidance.nice.org.uk/CG/Wave25/9/Consultation/Latest (accessed 19 March 2013)

104. Hornung T, Anwar GA. Faecal calprotectin: An audit to determine its clinical and cost effectiveness at North Tees and Hartlepool NHS Trust [abstract]. *Gut* 2011;60:A203.

105. Mindemark M, Larsson A. Ruling out IBD: estimation of the possible economic effects of pre-endoscopic screening with F-calprotectin. *Clinical Biochemistry* 2012;45:552-5.

106. Goldfarb NI, Pizzi LT, Fuhr JP, Jr., Salvador C, Sikirica V, Kornbluth A *et al*. Diagnosing Crohn's disease: an economic analysis comparing wireless capsule endoscopy with traditional diagnostic procedures. [Review] [69 refs]. *Disease Management* 2004;7:292-304.

107. Mascialino B, Hermansson L, Meister S, Mummert E. Cost-effectiveness in diagnostic tests: comparison of the IBD pre-endoscopic screening F-Calprotectin test versus serologic markers in the United Kingdom. Poster presented at ECHE (European Confrence on Health Economics, Zurich, Switzerland) 2012 http://eche2012.abstractsubmit.org/presentations/3407/ (accessed 7 March 2013)

108. Dubinsky MC, Johanson JF, Seidman EG, Ofman JJ. Suspected inflammatory bowel disease--the clinical and economic impact of competing diagnostic strategies. *Am J Gastroenterol* 2002;97:2333-42.

109. Ricci JF, Walters S, Ward S, Akehurst R. Quality of life of IBS patients in the United Kingdom compared with a general population sample [abstract]. *Gastroenterology* 2003;124:A396.

110. Bernklev T, Jahnsen J, Lygren I, Henriksen M, Vatn M, Moum B. Health-related quality of life in patients with inflammatory bowel disease measured with the short form-36: psychometric assessments and a comparison with general population norms. *Inflammatory Bowel Dis* 2005;11:909-18.

111. Brazier J, Roberts J, Tsuchiya A, Busschbach J. A comparison of the EQ-5D and SF-6D across seven patient groups. *Health Economics* 2004;13:873-84.

112. Bracco A, Jonsson B, Ricci JF, Drummond M, Nyhlin H. Economic evaluation of tegaserod vs. placebo in the treatment of patients with irritable bowel syndrome: an analysis of the TENOR study. *Value in Health* 2007;10:238-46.

113. Dibonaventura MD, Prior M, Prieto P, Fortea J. Burden of constipation-predominant irritable bowel syndrome (IBS-C) in France, Italy, and the United Kingdom. *Clin Exp Gastroenterol* 2012;5:203-12.

114. Pare P, Gray J, Lam S, Balshaw R, Khorasheh S, Barbeau M *et al*. Health-related quality of life, work productivity, and health care resource utilization of subjects with irritable bowel syndrome: baseline results from LOGIC (Longitudinal Outcomes Study of Gastrointestinal Symptoms in Canada), a naturalistic study. *Clinical Therapeutics* 2006;28:1726-35.

115. Puhan MA, Schunemann HJ, Wong E, Griffith L, Guyatt GH. The standard gamble showed better construct validity than the time trade-off. *Journal of Clinical Epidemiology* 2007;60:1029-33.

116. Konig HH, Ulshofer A, Gregor M, von TC, Reinshagen M, Adler G *et al*. Validation of the EuroQol questionnaire in patients with inflammatory bowel disease. *Eur J Gastroenterol Hepatol* 2002;14:1205-15.

117. Leidl R, Reitmeir P, Konig HH, Stark R. The performance of a value set for the EQ-5D based on experienced health states in patients with inflammatory bowel disease. *Value in Health* 2012;15:151-7.

118. Stark RG, Reitmeir P, Leidl R, Konig HH. Validity, reliability, and responsiveness of the EQ-5D in inflammatory bowel disease in Germany. *Inflammatory Bowel Dis* 2010;16:42-51.

119. Turunen P, Ashorn M, Auvinen A, Iltanen S, Huhtala H, Kolho KL. Long-term health outcomes in pediatric inflammatory bowel disease: a population-based study. *Inflammatory Bowel Dis* 2009;15:56-62.

120. Casellas F, Arenas JI, Baudet JS, Fabregas S, Garcia N, Gelabert J *et al*. Impairment of health-related quality of life in patients with inflammatory bowel disease: a Spanish multicenter study. *Inflammatory Bowel Dis* 2005;11:488-96.

121. Badia X, Roset M, Montserrat S, Herdman M, Segura A. [The Spanish version of EuroQol: a description and its applications. European Quality of Life scale]. *Med Clin (Barc)* 1999;112 Suppl 1:79-85.

122. Bassi A, Bodger K. Health state utilities & willingness-to-pay in inflammatory bowel disease (IBD) - a study of feasibility & validity [abstract]. *Gastroenterology* 2005;128:A555.

123. Arseneau KO, Cohn SM, Cominelli F, Connors AF, Jr. Cost-utility of initial medical management for Crohn's disease perianal fistulae. *Gastroenterology* 2001;120:1640-56.

124. Buxton MJ, Lacey LA, Feagan BG, Niecko T, Miller DW, Townsend RJ. Mapping from disease-specific measures to utility: an analysis of the relationships between the Inflammatory Bowel Disease Questionnaire and Crohn's Disease Activity Index in Crohn's disease and measures of utility. *Value in Health* 2007;10:214-20.

125. Benedini V, Caporaso N, Corazza GR, Rossi Z, Fornaciari G, Cottone M *et al*. Burden of Crohn's disease: economics and quality of life aspects in Italy. *Clinicoeconomics & Outcomes Research* 2012;4:209-18.

126. Casellas F, Rodrigo L, Nino P, Pantiga C, Riestra S, Malagelada JR. Sustained improvement of health-related quality of life in Crohn's disease patients treated with infliximab and azathioprine for 4 years. *Inflammatory Bowel Dis* 2007;13:1395-400.

127. Gregor JC, McDonald JW, Klar N, Wall R, Atkinson K, Lamba B *et al*. An evaluation of utility measurement in Crohn's disease. *Inflamm Bowel Dis* 1997;3:265-76.

128. Gibson PR, Weston AR, Shann A, Florin TH, Lawrance IC, Macrae FA *et al*. Relationship between disease severity, quality of life and health-care resource use in a cross-section of Australian patients with Crohn's disease. *Journal of Gastroenterology & Hepatology* 2007;22:1306-12.

129. Hill R, Lewindon P, Muir R, Grange I, Connor F, Ee L *et al*. Quality of life in children with Crohn disease. *Journal of Pediatric Gastroenterology & Nutrition* 2010;51:35-40.

130. Connolly MP, Poole CD, Currie CJ, Marteau P, Nielsen SK. Quality of life improvements attributed to combination therapy with oral and topical mesalazine in mild-to-moderately active ulcerative colitis. *Digestion* 2009;80:241-6.

131. Bryan S, Andronis L, Hyde C, Connock M, Fry-Smith A, Wang D. Infliximab for acute exacerbations of ulcerative colitis. 2008 http://www.hta.ac.uk/erg/reports/1730.pdf (accessed 29 March 2013)

132. Arseneau KO, Sultan S, Provenzale DT, Onken J, Bickston SJ, Foley E *et al*. Do patient preferences influence decisions on treatment for patients with steroid-refractory ulcerative colitis? *Clin Gastroenterol Hepatol* 2006;4:1135-42.

133. Feagan BG, Reinisch W, Rutgeerts P, Sandborn WJ, Yan S, Eisenberg D *et al*. The effects of infliximab therapy on health-related quality of life in ulcerative colitis patients. *American Journal of Gastroenterology* 2007;102:794-802.

134. Waljee AK, Higgins PD, Waljee JF, Tujios SR, Saxena A, Brown LK *et al*. Perceived and actual quality of life with ulcerative colitis: a comparison of medically and surgically treated patients. *American Journal of Gastroenterology* 2011;106:794-9.

135. Poole CD, Connolly MP, Nielsen SK, Currie CJ, Marteau P. A comparison of physician-rated disease severity and patient reported outcomes in mild to moderately active ulcerative colitis. *J Crohns Colitis* 2010;4:275-82.

136. Baudet JS, Diaz-Bethencourt D, Aviles J, Aguirre-Jaime A. Minor adverse events of colonoscopy on ambulatory patients: the impact of moderate sedation. *Eur J Gastroenterol Hepatol* 2009;21:656-61.

137. de Jonge V, Nicolaas JS, van BO, Brouwer JT, Stolk MF, Tang TJ *et al*. The incidence of 30-day adverse events after colonoscopy among outpatients in the Netherlands. *American Journal of Gastroenterology* 2012;107:878-84.

138. Dominitz JA, Provenzale D. Patient preferences and quality of life associated with colorectal cancer screening. *Am J Gastroenterol* 1997;92:2171-8.

139. Niv Y, Bogolavski I, Ilani S, Avni I, Gal E, Vilkin A *et al*. Impact of colonoscopy on quality of life. *Eur J Gastroenterol Hepatol* 2012;24:781-6.

140. Spiegel BM, Gralnek IM, Bolus R, Chang L, Dulai GS, Naliboff B *et al*. Is a negative colonoscopy associated with reassurance or improved health-related quality of life in irritable bowel syndrome? *Gastrointest Endosc* 2005;62:892-9.

141. Warren JL, Klabunde CN, Mariotto AB, Meekins A, Topor M, Brown ML *et al*. Adverse events after outpatient colonoscopy in the Medicare population. *Annals of Internal Medicine* 2009;150:849-57.

142. Levin TR, Zhao W, Conell C, Seeff LC, Manninen DL, Shapiro JA *et al*. Complications of colonoscopy in an integrated health care delivery system. *Ann Intern Med* 2006;145:880-6.

143. National Institute for Health and Clinical Excellence. Colonoscopic surveillance for prevention of colorectal cancer in people with ulcerative colitis, Crohn's disease or adenomas (CG118). 2011 http://guidance.nice.org.uk/CG118 (accessed 11 March 2013)

144. Tappenden P, Eggington S, Nixon R, Chilcott J, Sakai H, Karnon J. Colorectal cancer screening options appraisal: Report to the English Bowel Cancer Screening Working Group. 2004 http://www.cancerscreening.nhs.uk/bowel/scharr.pdf (accessed 11 March 2013)

145. Atkin WS, Edwards R, Kralj-Hans I, Wooldrage K, Hart AR, Northover JM *et al*. Once-only flexible sigmoidoscopy screening in prevention of colorectal cancer: a multicentre randomised controlled trial. *Lancet* 2010;375:1624-33.

146. Atkin WS, Cook CF, Cuzick J, Edwards R, Northover JM, Wardle J. Single flexible sigmoidoscopy screening to prevent colorectal cancer: baseline findings of a UK multicentre randomised trial. *Lancet* 2002;359:1291-300.

147. Gatto NM, Frucht H, Sundararajan V, Jacobson JS, Grann VR, Neugut AI. Risk of perforation after colonoscopy and sigmoidoscopy: a population-based study. *J Natl Cancer Inst* 2003;95:230-6.

148. Mearin F, Roset M, Badia X, Balboa A, Baro E, Ponce J *et al*. Splitting irritable bowel syndrome: from original Rome to Rome II criteria. *Am J Gastroenterol* 2004;99:122-30.

149. Moroni F, Winter JW, Morris AJ, Gaya DR. What is the clinical relevance of a mildly elevated faecal calprotectin detected in new referrals to the gastroenterology clinic? *Gut* 2012;61:A78-A79.

150. Zayyat R, Appleby RN, Logan RPH. Can we improve the negative predictive value of faecal calprotectin for the diagnosis of ibs in primary care? [abstract]. *Gut* 2011;60:A49-A50.

151. Mohammed N, Smale S. Positive calprotectin but negative investigations-what next? *Gut* 2012;61:A236.

152. Demir OM, Ahmed Z, Logan RPH. Optimising the use of faecal calprotectin for early diagnosis of IBD in primary care. *Journal of Crohn's and Colitis* 2013;7:S8-S9.

153. Koulaouzidis A, Douglas S, Rogers MA, Arnott ID, Plevris JN. Fecal calprotectin: a selection tool for small bowel capsule endoscopy in suspected IBD with prior negative bi-directional endoscopy. *Scand J Gastroenterol* 2011;46:561-6.

154. Farrokhyar F, Marshall JK, Easterbrook B, Irvine EJ. Functional gastrointestinal disorders and mood disorders in patients with inactive inflammatory bowel disease: prevalence and impact on health. *Inflamm Bowel Dis* 2006;12:38-46.

155. Keohane J, O'Mahony C, O'Mahony L, O'Mahony S, Quigley EM, Shanahan F. Irritable bowel syndrome-type symptoms in patients with inflammatory bowel disease: a real

association or reflection of occult inflammation? *American Journal of Gastroenterology* 2010;105:1788-94.

156. Nurmi E, Haapamaki J, Paavilainen E, Rantanen A, Hillila M, Arkkila P. The burden of inflammatory bowel disease on health care utilization and quality of life. *Scand J Gastroenterol* 2013;48:51-7.

157. Jelsness-Jorgensen LP, Bernklev T, Moum B. Calprotectin Is a Useful Tool in Distinguishing Coexisting Irritable Bowel-Like Symptoms from That of Occult Inflammation among Inflammatory Bowel Disease Patients in Remission. *Gastroenterol Res Pract* 2013;2013:620707.

158. Canani RB, Terrin G, Rapacciuolo L, Miele E, Siani MC, Puzone C *et al*. Faecal calprotectin as reliable non-invasive marker to assess the severity of mucosal inflammation in children with inflammatory bowel disease. *Digestive & Liver Disease* 2008;40:547-53.

159. Fagerberg UL, Loof L, Lindholm J, Hansson LO, Finkel Y. Fecal calprotectin: a quantitative marker of colonic inflammation in children with inflammatory bowel disease. *Journal of Pediatric Gastroenterology & Nutrition* 2007;45:414-20.

160. van Rheenen PF. Role of fecal calprotectin testing to predict relapse in teenagers with inflammatory bowel disease who report full disease control. *Inflamm Bowel Dis* 2012.

161. Panaccione R, Hibi T, Peyrin-Biroulet L, Schreiber S. Implementing changes in clinical practice to improve the management of Crohn's disease. *J Crohns Colitis* 2012;6 Suppl 2:S235-S242.

162. D'Haens G, Ferrante M, Vermeire S, Baert F, Noman M, Moortgat L *et al*. Fecal calprotectin is a surrogate marker for endoscopic lesions in inflammatory bowel disease. *Inflamm Bowel Dis* 2012;Article first published online: 16 FEB 2012.

163. Lees C, Clark A, Walkden A, Satsangi J, Arnott I. Clinical utility of faecal calprotectin in diagnosing IBD during first presentation to the gastroenterology clinic: A novel investigative algorithm [abstract]. *J Crohn's Colitis Suppl* 2010;4:32.

164. National Institute for Health and Clinical Excellence. SeHCAT (Tauroselcholic [75Selenium] acid) for the investigation of bile acid malabsorption (BAM) and measurement of bile acid pool loss - Diagnostics guidance, DG7. 2012 http://guidance.nice.org.uk/DG7 (accessed 7 March 2013)

165. Alrubaiy L, Malik A, Rees I, Bowen D. Usefulness of Fecal Calprotectin in Clinical Practice in a District General Hospital. *Inflammatory Bowel Dis* 2012;18:S53-S54.

166. Taylor N, Hills E, Sheen C, Al-Bahrani A, Grellier L. An audit of faecal calprotectin testing in suspected inflammatory bowel disease in the under 45's. *Journal of Crohn's and Colitis* 2013;7:S131.

167. Foell D, Wittkowski H, Vogl T, Roth J. S100 proteins expressed in phagocytes: a novel group of damage-associated molecular pattern molecules. *J Leukoc Biol* 2007;81:28-37.

168. Kaiser T, Langhorst J, Wittkowski H, Becker K, Friedrich AW, Rueffer A *et al*. Faecal S100A12 as a non-invasive marker distinguishing inflammatory bowel disease from irritable bowel syndrome. *Gut* 2007;56:1706-13.

169. Turner D, Leach ST, Mack D, Uusoue K, McLernon R, Hyams J *et al*. Faecal calprotectin, lactoferrin, M2-pyruvate kinase and S100A12 in severe ulcerative colitis: a prospective multicentre comparison of predicting outcomes and monitoring response. *Gut* 2010;59:1207-12.

170. Roseth AG, Kristinsson J, Fagerhol MK, Schjonsby H, Aadland E, Nygaard K *et al*. Faecal calprotectin: a novel test for the diagnosis of colorectal cancer? *Scand J Gastroenterol* 1993;28:1073-6.

171. Tibble JA, Sigthorsson G, Foster R, Scott D, Fagerhol MK, Roseth A *et al*. High prevalence of NSAID enteropathy as shown by a simple faecal test. *Gut* 1999;45:362-6.

172. Mooiweer E, Fidder HH, Van Erpecum KJ, Siersema PD, Laheij RJ, Oldenburg B. Determination of faecal haemoglobin is equally effective as faecal calprotectin in identifying inflammatory bowel disease patients with active endoscopic inflammation. *Journal of Crohn's and Colitis* 2013;7:S114.

173. Sherwood RA. Faecal markers of gastrointestinal inflammation. *J Clin Pathol* 2012;65:981-5.

174. Sutherland AD, Gearry RB, Frizelle FA. Review of fecal biomarkers in inflammatory bowel disease. [Review] [56 refs]. *Diseases of the Colon & Rectum* 2008;51:1283-91.

175. D'Inca R, Dal PE, Di L, V, Ferronato A, Fries W, Vettorato MG *et al*. Calprotectin and lactoferrin in the assessment of intestinal inflammation and organic disease. *Int J Colorectal Dis* 2007;22:429-37.

176. Joishy M, Davies I, Ahmed M, Wassel J, Davies K, Sayers A *et al*. Fecal calprotectin and lactoferrin as noninvasive markers of pediatric inflammatory bowel disease. *Journal of Pediatric Gastroenterology & Nutrition* 2009;48:48-54.

177. Nancey S, Boschetti G, Moussata D, Cotte E, Peyras J, Cuerq C *et al*. Neopterin is a novel reliable fecal marker as accurate as calprotectin for predicting endoscopic disease activity in patients with inflammatory bowel diseases. *Inflamm Bowel Dis* 2013;19:1043-52.

178. Arasaradnam RP, Ouaret N, Thomas MG, Quraishi N, Heatherington E, Nwokolo CU *et al*. A novel tool for noninvasive diagnosis and tracking of patients with inflammatory bowel disease. *Inflamm Bowel Dis* 2013;19:999-1003.

179. University of British Columbia. FOCUS: The Future of Fecal Calprotectin Utility Study for the Diagnosis and Management of Inflammatory Bowel Disease (IBD). 2013 http://clinicaltrials.gov/ct2/results?term=NCT01676324&Search=Search (accessed 30 March 2013)

180. Editors: Colledge, N. R., Walker, B. R., and Ralston, S. H. Davidson's principles and practice of medicine. 21st ed. 2010. Edinburgh, New York, Churchill Livingstone/Elsevier.

181. Bruzzese E, Raia V, Gaudiello G, Polito G, Buccigrossi V, Formicola V *et al*. Intestinal inflammation is a frequent feature of cystic fibrosis and is reduced by probiotic administration. *Alimentary Pharmacology & Therapeutics* 2004;20:813-9.

182. Bunn SK, Bisset WM, Main MJ, Golden BE. Fecal calprotectin as a measure of disease activity in childhood inflammatory bowel disease. *Journal of Pediatric Gastroenterology & Nutrition* 2001;32:171-7.

183. Canani RB, Passaro M, Puzone C. Diagnostic value of faecal calprotectin in paediatric age. *Med Bambino* 2009;28:239-42.

184. Costa F, Mumolo MG, Bellini M, Romano MR, Ceccarelli L, Arpe P *et al*. Role of faecal calprotectin as non-invasive marker of intestinal inflammation. *Digestive & Liver Disease* 2003;35:642-7.

185. Eder P, Stawczyk-Eder K, Krela-Kazmierczak I, Linke K. Clinical utility of the assessment of fecal calprotectin in Lesniowski-Crohn's disease. *Pol Arch Med Wewn* 2008;118:622-6.

186. El Nuamani N, El-Gendi H, Zaki S, Elamin H, Tawfik M, Al-Sheshtawy F. Evaluation of the Diagnostic Yield of Faecal Calprotectin in Egyptian Children with Chronic Diarrhoea. *Arab J Gastroenterol* 2007;8:132-5.

187. Elkjaer M, Burisch J, Voxen H, V, Deibjerg KB, Slott Jensen JK, Munkholm P. A new rapid home test for faecal calprotectin in ulcerative colitis. *Alimentary Pharmacology & Therapeutics* 2010;31:323-30.

188. Erbayrak M, Turkay C, Eraslan E, Cetinkaya H, Kasapoglu B, Bektas M. The role of fecal calprotectin in investigating inflammatory bowel diseases. *Clinics (Sao Paulo, Brazil)* 2009;64:421-5.

189. Flagstad G, Helgeland H, Markestad T. Faecal calprotectin concentrations in children with functional gastrointestinal disorders diagnosed according to the Pediatric Rome III criteria. *Acta Paediatrica* 2010;99:734-7.

190. Grogan JL, Casson DH, Terry A, Burdge GC, El-Matary W, Dalzell AM. Enteral feeding therapy for newly diagnosed pediatric Crohn's disease: a double-blind randomized controlled trial with two years follow-up. *Inflamm Bowel Dis* 2012;18:246-53.

191. Guo H-B, Cao J-B, Wang Z-H, Li H-R. Fecal calprotectin in differential diagnosis between irritable bowel syndrome and inflammatory bowel disease. *World Chin J Dig* 2009;17:1152-5.

192. Jensen MD, Kjeldsen J, Nathan T. Fecal calprotectin is equally sensitive in Crohn's disease affecting the small bowel and colon. *Scand J Gastroenterol* 2011;46:694-700.

193. Kobelska-Dubiel N, Igny-acs I, Krauss H, Cichy W, Kobelski M. [Fecal calprotectin as an inflammatory marker in inflammatory bowel diseases in children] [Polish]. *Pediatr Wspolczesna* 2007;9:172-5.

194. Langhorst J, Elsenbruch S, Koelzer J, Rueffer A, Michalsen A, Dobos GJ. Noninvasive markers in the assessment of intestinal inflammation in inflammatory bowel diseases: performance of fecal lactoferrin, calprotectin, and PMN-elastase, CRP, and clinical indices. *American Journal of Gastroenterology* 2008;103:162-9.

195. Meucci G, D'Inca R, Maieron R, Orzes N, Vecchi M, Visentini D *et al*. Diagnostic value of faecal calprotectin in unselected outpatients referred for colonoscopy: A multicenter prospective study. *Dig Liver Dis* 2010;42:191-5.

196. Olafsdottir E, Aksnes L, Fluge G, Berstad A. Faecal calprotectin levels in infants with infantile colic, healthy infants, children with inflammatory bowel disease, children with recurrent abdominal pain and healthy children. *Acta Paediatrica* 2002;91:45-50.

197. Perminow G, Beisner J, Koslowski M, Lyckander LG, Stange E, Vatn MH *et al*. Defective paneth cell-mediated host defense in pediatric ileal Crohn's disease. *American Journal of Gastroenterology* 2010;105:452-9.

198. Quail MA, Russell RK, Van Limbergen JE, Rogers P, Drummond HE, Wilson DC *et al*. Fecal calprotectin complements routine laboratory investigations in diagnosing childhood inflammatory bowel disease. *Inflammatory Bowel Dis* 2009;15:756-9.

199. Ricanek P, Brackmann S, Perminow G, Lyckander LG, Sponheim J, Holme O *et al*. Evaluation of disease activity in IBD at the time of diagnosis by the use of clinical, biochemical, and fecal markers. *Scand J Gastroenterol* 2011;46:1081-91.

200. Roseth AG, Aadland E, Jahnsen J, Raknerud N. Assessment of disease activity in ulcerative colitis by faecal calprotectin, a novel granulocyte marker protein. *Digestion* 1997;58:176-80.

201. Roseth AG, Schmidt PN, Fagerhol MK. Correlation between faecal excretion of indium-111-labelled granulocytes and calprotectin, a granulocyte marker protein, in patients with inflammatory bowel disease. *Scand J Gastroenterol* 1999;34:50-4.

202. Schoepfer AM, Beglinger C, Straumann A, Trummler M, Renzulli P, Seibold F. Ulcerative colitis: correlation of the Rachmilewitz endoscopic activity index with fecal calprotectin, clinical activity, C-reactive protein, and blood leukocytes. *Inflammatory Bowel Dis* 2009;15:1851-8.

203. Schoepfer AM, Beglinger C, Straumann A, Trummler M, Vavricka SR, Bruegger LE *et al*. Fecal calprotectin correlates more closely with the Simple Endoscopic Score for Crohn's disease (SES-CD) than CRP, blood leukocytes, and the CDAI. *American Journal of Gastroenterology* 2010;105:162-9.

204. Shastri YM, Povse N, Stein J. A prospective comparative study for new rapid bedside fecal calprotectin test with an established ELISA to assess intestinal inflammation. *Clin Lab* 2009;55:53-5.

205. Shulman RJ, Eakin MN, Czyzewski DI, Jarrett M, Ou CN. Increased gastrointestinal permeability and gut inflammation in children with functional abdominal pain and irritable bowel syndrome. *Journal of Pediatrics* 2008;153:646-50.

206. Sidhu R, Wilson P, Wright A, Yau CWH, D'Cruz FA, Foye L *et al*. Faecal lactoferrin - a novel test to differentiate between the irritable and inflamed bowel? *Alimentary Pharmacology & Therapeutics* 2010;31:1365-70.

207. Sipponen T, Karkkainen P, Savilahti E, Kolho KL, Nuutinen H, Turunen U *et al*. Correlation of faecal calprotectin and lactoferrin with an endoscopic score for Crohn's disease and histological findings. *Alimentary Pharmacology & Therapeutics* 2008;28:1221-9.

208. Sipponen T, Savilahti E, Kolho KL, Nuutinen H, Turunen U, Farkkila M. Crohn's disease activity assessed by fecal calprotectin and lactoferrin: correlation with Crohn's disease activity index and endoscopic findings. *Inflammatory Bowel Dis* 2008;14:40-6.

209. Sipponen T, Haapamaki J, Savilahti E, Alfthan H, Hamalainen E, Rautiainen H *et al*. Fecal calprotectin and S100A12 have low utility in prediction of small bowel Crohn's disease detected by wireless capsule endoscopy. *Scand J Gastroenterol* 2012;47:778-84.

210. Summerton CB, Longlands MG, Wiener K, Shreeve DR. Faecal calprotectin: a marker of inflammation throughout the intestinal tract. *Eur J Gastroenterol Hepatol* 2002;14:841-5.

211. Tibble J, Teahon K, Thjodleifsson B, Roseth A, Sigthorsson G, Bridger S *et al*. A simple method for assessing intestinal inflammation in Crohn's disease. *Gut* 2000;47:506-13.

212. Uslu N, Baysoy G, Balamtekin N, Hizal G, Demir H, Saltik-Temizel IN *et al*. [A noninvasive marker for pediatric inflammatory bowel disease: Fecal calprotectin] {Turkish]. *Cocuk Sagligi Hast Derg* 2011;54:22-7.

213. Wassell J, Dolwani S, Metzner M, Losty H, Hawthorne A. Faecal calprotectin: a new marker for Crohn's disease? *Ann Clin Biochem* 2004;41:3-2.

214. Xiang JY, Ouyang Q, Li GD, Xiao NP. Clinical value of fecal calprotectin in determining disease activity of ulcerative colitis. *World Journal of Gastroenterology* 2008;14:53-7.

Appendices

[source: Davidson's Principles and Practice of Medicine 21st Ed[180]]

Appendix 1 Comparison of ulcerative colitis, Crohn's disease, IBS and coeliac disease

Table 67. Comparison between ulcerative colitis, Crohn's disease, IBS and coeliac disease

	Ulcerative Colitis	Crohn's disease	IBS	Coeliac disease
Age	Any	Any	Any, but more in young	Any age. Adults peak onset in third or fourth decade
Gender	M=F	M=F	Female preponderance	In adults, females are affected twice as often as males
Population distribution	Incidence in Western countries: 10 to 20 per 100,000 Prevalence in Western countries: 100 to 200 per 100,000	Incidence in Western countries: 5 to 10 per 100,000 Prevalence in Western countries: 50 to 100 per 100,000	About 20% but only 10% consult GPs.	World-wide but more common in northern Europe Prevalence in the UK approx. 1%, but 50% of them are asymptomatic (include both undiagnosed 'silent' cases and 'latent' cases, who later develop coeliac disease)
Ethnic group	Any	Any; more common in Ashkenazi Jews	Not reported	Not mentioned
Genetic factors	*HLA-DR103* associated with severe disease	*CARD 15/NOD-2* mutations predispose	Not reported	Genetically susceptible individuals intolerant to wheat gluten and similar proteins found in rye, barley and, to oats; Associated with other HLA-linked autoimmune disorders and with certain other diseases
Risk factors	More common in non-/ex-smokers; Appendicetomy protects	More common in smokers (RR = 3)	History of psychological stress.	Not mentioned as risk factors but coeliac disease associated with other HLA-linked automimmune disorders and with certain other diseases like IDDM, thyroid disease, primary biliary cirrhosis, IBD, Sjogren's syndrome, IgA def, pernicious anaemia,

				sarcoidosis, myasthenia gravis, down's syndrome etc
Diagnosis	Clinical confirmed by biopsy	Biopsy	Clinical. Diagnosis supported by symptoms for more than 6 months; worsened by stress; FBC and ESR normal	Endoscopic small bowel biopsy is the gold standard (villous atrophy); IgA anti-endomysial antibodies by immunofluorescence; Haematology (micro and macrocytic anaemia)and biochemistry (low ca, mg, total protein, albumin and Vit D);
Anatomical distribution	Colon only; begins at anorectal margin with variable proximal extension Proctitis (rectum); proctosigmoiditis (retum and sigmoid colon); pancolitis (whole colon)	Any part of gastrointestinal tract; perianal disease common; patchy distribution – 'skip lesions' Sites involved (in order of frequency): terminal ileum and right side of colon, colon alone, terminal ileum alone, ileum and jejunum	Colon.	Small bowel
Extraintestinal manifestations	Common	Common	Associated with other conditions such as dysmenorrhoaea, non-ulcer dyspepsia, "fibromyalgia".	Common
Presentation	Bloody diarrhoea *Proctitis* – rectal bleeding, mucus discharge, tenesmus *Proctosigmoiditis* – bloody diarrhoea with mucus; some develop fever, lethargy and abdominal discomfort *Extensive pancolitis* – bloody diarrhoea with passage of mucus. Severe case – anorexia, malaise, weight loss and abdominal pain,	Variable; pain, diarrhoea, weight loss all common *Ileal Crohn's disease:* there may be subacute or even acute intestinal obstruction. Diarrhoea- watery but no blood or mucus *Crohn's colitis:* identical to UC but rectum spared and presence of perianal disease. Many presents with symptoms of both small bowel and	Recurrent colicky abdominal pain or cramping, relieved by defecation. Abdominal distension. Episodes of diarrhoea but can have more of a constipation pattern. Patients well, no weight loss.	Depends on age of onset Infancy: diarrhoea, malabsorption and failure to thrive – symptoms starts after weaning on to cereals Older children: non-specific features like delayed growth; malnutrition, mild abdominal distension; growth and pubertal delay Adults: Highly

	patient is toxic with fever, tachycardia and signs of peritoneal inflammation	colonic disease. In few, isolated perianal disease, vomiting from jejunal strictures and severe oral ulceration		variable, depending on the severity and extent of small bowel involvement. Some florid malabsorption, others nonspecific symptoms such as tiredness, weight loss, folate or iron def anaemia. Oral ulceration, dyspepsia and bloating; mild undernutrition and increased risk of osteoporosis
Histology	Inflammation limited to mucosa; crypt distortion, cryptitis, crypt abscesses, loss of goblet cells	Submucosal or transmural inflammation common; deep fissuring ulcers, fistulas; patchy changes; granulomas	Normal	Subtotal villous atrophy. Sometime villous appears normal but there may be excess numbers of intra-epithelial lymphocytes

Appendix 2. Search strategy

Calprotectin - diagnostic studies and economics

Medline (Ovid) (1946 to September 2012)

1. exp Inflammatory Bowel Diseases/di [Diagnosis]
2. exp Irritable Bowel Syndrome/di [Diagnosis]
3. crohn's disease.tw.
4. ulcerative colitis.tw.
5. inflammatory bowel disease*.tw.
6. irritable bowel syndrome*.tw.
7. (IBS or IBD).tw.
8. 1 or 2 or 3 or 4 or 5 or 6 or 7
9. calprotectin.tw.
10. 8 and 9

Embase (Ovid) (1980 to September 2012)

1. crohn's disease.tw.
2. ulcerative colitis.tw.
3. inflammatory bowel disease*.tw.
4. irritable bowel syndrome*.tw.
5. (IBS or IBD).tw.
6. calprotectin.tw.
7. 1 or 2 or 3 or 4 or 5
8. 6 and 7
9. exp *Crohn disease/di [Diagnosis]
10. exp *ulcerative colitis/di [Diagnosis]
11. 9 or 10
12. 6 and 11
13. 8 or 12
14. (fecal or faecal).tw.
15. 13 and 14

Auto-alerts

Ran auto-alerts of the above searches in Medline and Embase from September 2012 to March 2013 for studies added subsequent to the initial searches.

Cochrane library – all sections, Sept 2012

Search terms**:** calprotectin and (inflammatory bowel disease* or irritable bowel syndrome or crohn's disease or ulcerative colitis)

Web of Science – Science Citation Index, Conference Proceedings Citation Index 1980 to September 2012

Search terms*:* calprotectin and (inflammatory bowel disease* or irritable bowel syndrome or crohn's disease or ulcerative colitis)

Cost effectiveness searches

Ovid MEDLINE 1996 to October 2012; Embase 1996- October 2012

1. exp Economics/
2. Health Status/
3. exp "Quality of Life"/
4. exp Quality-Adjusted Life Years/
5. exp Patient Satisfaction/
6. (pharmacoeconomic* or pharmaco-economic* or economic* or cost-effective* or cost-benefit*).tw.
7. (health state* or health status).tw.
8. (qaly* or utilit* or EQ5D or EQ-5D or euroqol or euro-qol or SF-36 or SF36 or SF-6D or HUI).tw.
9. (markov or time trade off or standard gamble or hrql or hrqol or disabilit* or disutilit*).tw.
10. (quality adj2 life).tw.
11. (decision adj2 model).tw.
12. (visual analog* scale* or discrete choice experiment* or health* year* equivalen*).tw.
13. ("resource use" or resource utili?ation).tw.
14. (well-being or wellbeing or satisfaction).ti.
15. 1 or 2 or 3 or 4 or 5 or 6 or 7 or 8 or 9 or 10 or 11 or 12 or 13
16. exp Inflammatory Bowel Diseases/ec, px [Economics, Psychology]
17. exp Irritable Bowel Syndrome/ec, px [Economics, Psychology]
18. (inflammatory bowel disease* or crohn* disease or ulcerative colitis).ti.
19. irritable bowel.m_titl.
20. 17 or 18 or 19
21. 15 and 20
22. limit 21 to english language
23. limit 22 to yr="1996 -Current"

Cochrane Library, Economic Evaluations Database, Issue 4 of 4, Oct 2012

Search terms: (inflammatory bowel or irritable bowel or crohn* or ulcerative colitis)

Other searches for calprotectin

- Searched the website of the journal *Gut*
- Searched ECCO (European Crohn's and Colitis Organisation) 2012 and 2013 Congress abstracts
- Checked reference lists of previous systematic reviews
- Personal communication with experts for unpublished data

Searches for adverse effects of colonoscopy

Ovid Medline 1946 to February2013

1. exp *Colonoscopy/ae [Adverse Effects]
2. (Colonoscopy or Sigmoidoscopy).m_titl.
3. (perforation* or peforated or complication*).tw.
4. 2 and 3
5. 1 or 4
6. limit 5 to english language
7. (case reports or comment or letter).pt.
8. 6 not 7
9. colonoscopy.m_titl.
10. 8 and 9

PubMed all database up to Feb 2013

Search terms: (colonoscopy and (perforat* or adverse or complication* or risk)) in title field.

Natural history/progression of IBD

1. exp Inflammatory Bowel Diseases/
2. (inflammatory bowel disease* or crohn* disease or ulcerative colitis).ti.
3. 1 or 2
4. (natural history or (disease adj course) or (clinical course) or progression or (disease adj2 progress*)).tw.
5. exp Disease Progression/
6. 4 or 5
7. 3 and 6
8. limit 7 to english language

Ovid MEDLINE1946 to October 2012

1. (inflammatory bowel disease* or crohn* disease or ulcerative colitis).ti.
2. (natural history or (disease adj course) or (clinical course) or progression or (disease adj2 progress*)).ti.
3. 1 and 2
4. limit 3 to english language

Research in progress

Included only open studies and excluded studies with unknown status

1. ClinicalTrials.gov
2. Current Controlled Trials
3. UK Clinical Trials Gateway
4. UK Clinical Research Network Study Portfolio
5. EU Clinical Trials Register website
6. EUDRACT European Clinical Trials Database
7. WHO (World Health Organization) Clinical Trials Search Portal

Flow Diagram

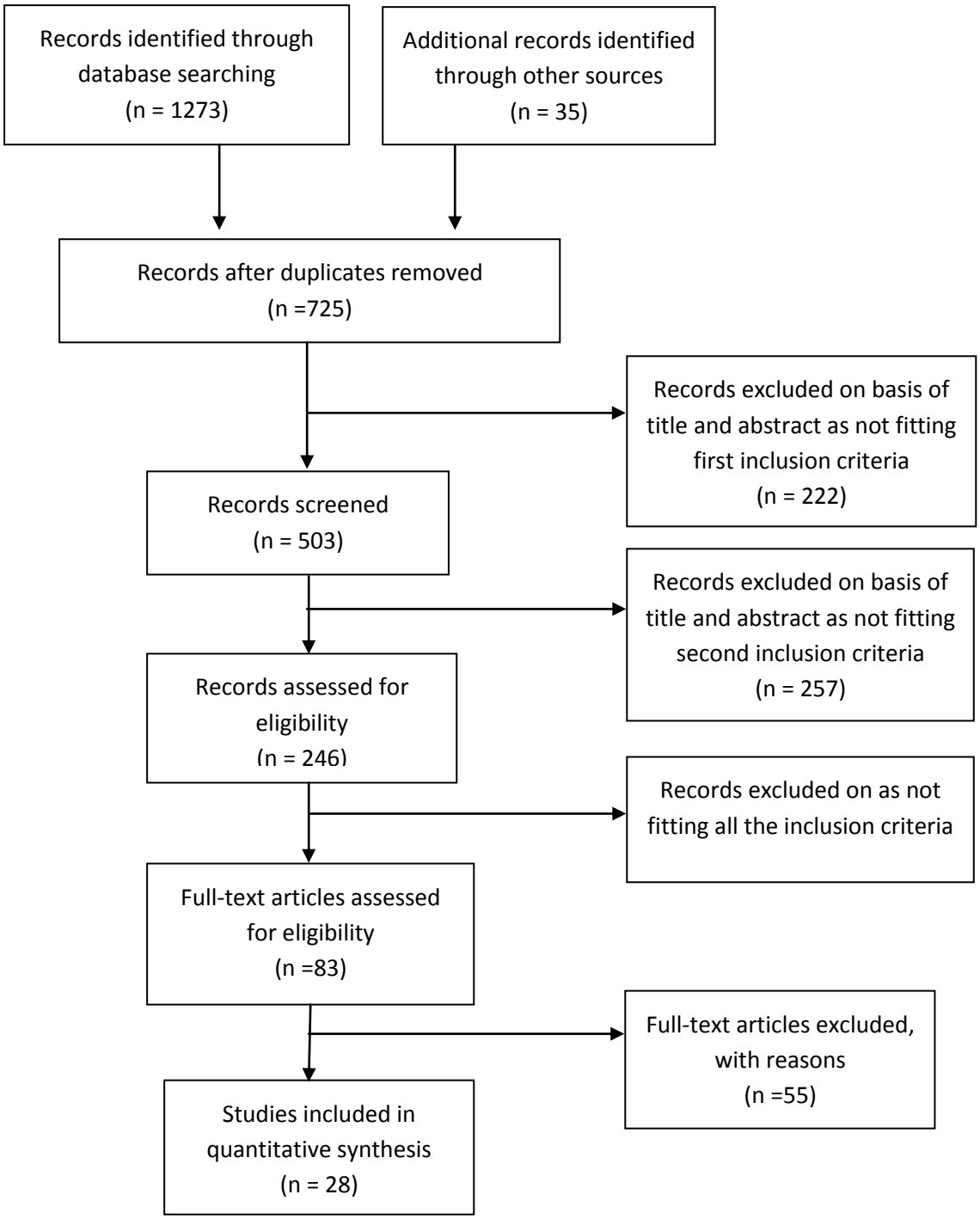


Table 68. Reasons for exclusion

First author and year	Reason for exclusion
Aomatsu 2011[59]	Previously diagnosed - and does not say how long since diagnosis
Bremner 2005[84]	Only 12/43 (28%) were newly diagnosed
Bruzzese 2004[181]	Not enough detail about the 15 patients with active IBD
Bunn 2001[63]	Only 9/22 (41%) are newly diagnosed - remainder under review on treatment [from data in Table 1]
Bunn 2001[182]	Not newly diagnosed**,** 13/22 (59%) under review on treatment
Canani 2004[183]	Could not derive a 2x2 table; includes patients in remision (proportion not reported)
Canani 2008[158]	States that patients had previously established diagnosis of IBD
Costa 2003[184]	Does not state what % of patients not previously diagnosed - first para p.645 describes patients with clinically active disease and those with quiescent disease i.e.mixture of patients
D'Inca 2007[175]	Patients with IBD had known diagnosis - referred for active disease or surveillance
Dolci 2012[90]	Letter - no information on patients - could not get 2x2 data - laboratory based setting
Eder 2008[185]	Confirmed diagnosis of CD - mean duration 5 years
El Nuamani 2007[186]	Requested full text but library unable to locate it
Elkjaer 2010 [187]	Patients with known CD - but maybe useful to compare POCT vs ELISA?
Erbayrak 2009[188]	Previously diagnosed - mean disease period 5 years
Fagerberg 2007[159]	Comprises a mixture of children with suspected or previously confirmed IBD - but does not give us the numbers of each
Flagstad 2010[189]	Spectrum bias as only looked at children with functional GI disorders - organic disease was excluded
Grogan 2012[190]	Spectrum bias - excluded children with only large bowel disease
Guo 2009[191]	Chinese language. Appeared from abstract that patients had a known diagnosis.
Hessels 2012 [92]	Comprised a mixture of patients with suspected (40/85) or relapse of IBD
Hornung 2011[104]	Health economics study, no 2x2 data available
Jensen 2011[192]	Possible spectrum bias; also, does not give the diagnosis of 43 of the 83 patients who were not CD i.e. we don't know how many of them had inflammatory vs non-inflammatory.
Joishy 2009[176]	Includes new cases and those in relapse, but doesn't say how many of each
Keohane 2010 [155]	Patients only included if in clinical remission
Kobelska-Dubiel 2007[193]	Polish language- not able to get translation.
Komraus 2012[85]	Patients had a known diagnosis of various types of IBD; could not get data for a 2x2 table
Koulaouzidis 2011[153]	Spectrum bias - all patients had previously had negative bidirectional endoscopies but continuining suspicion of CD and referred for small bowel capsule endoscopy
Langhorts 2008[194]	Only patients with previously diagnosed IBD were included
Meucci 2010[195]	Not all new onset. High proportion of IBD patients had clinically quiescent IBD - did not report FC data for active IBD separately - so could not derive 2x2 table
Mindemark 2012[105]	Health economics paper - no 2x2 data avaiable
Moum 2010[37]	Article looked at reproducibility of tests - no 2x2 data available

Olafsdottir 2002[196]	Majority of patients not have the reference standard of colonoscopy
Perminow 2010[197]	Not enough detail to extract sensitivity and specificity data
Quail 2009[198]	Measured at diagnosis but not enough detail reported to derive data for a 2x2 table
Ricanek 2011[199]	Does not give the manufacturer of the test and does not give a cut-off value for the test - does not report enough data to construct a 2x2 table
Roseth 1997[200]	Most patients were previously diagnosed - only 9/62 =15% underwent first diagnostic exam- median disease duration is 5 years
Roseth 1999[201]	Patients appeared to be in remission and were receiving maintenance treatment only
Schoepfer 2007[66]	Not newly presenting patients in a differential diagnosis situation - 19 of 24 CD patients in remission - so had no symptoms.
Schoepfer 2009[202]	Previously confirmed diagnosis of UC referred for colonoscopy'
Schoepfer 2010[203]	Previously confirmed diagnosis of CD referred for colonoscopy'
Shastri 2009[204]	Poster - not enough details about patients or data for 2x2 tables
Shaoul 2012[83]	Not enough data for a 2x2 table
Shulman2008[205]	Only includes children with functional abdominal pain (IBS)
Sidhu 2010[206]	Includes patients with known diagnosis -about 44% active disease and 66% inactive disease
Sipponen 2008[207]	Includes patients with an established CD diagnosis
Sipponen 2008[208]	Not newly diagnosed - patients referrred for ileocolonoscopy had a disease duration of mean 9.2 years
Sipponen 2012[209]	Time between index test and reference test greater than 3 months. Spectrum bias - other tests had already been done. Small bowel only.
Summerton 2002[210]	Most had a known diagnosis, so not new. No data on IBS or IBD in relation to FC cut-off levels
Sydora 2012[93]	Patients have had a known diagnosis prior to the study
Tibble 2000[211]	Patients appeared to be a sub-group of those included in Tibble 2002
Tomkins 2012[99]	A mixture of previously diagnosed and newly presenting patients but numbers of each not known - personal communication
Uslu 2011[212]	Turkish language - not able to get translation
Vestergaard 2008[94]	Patients not newly diagnosed - aim of the study was to compare RAPID and ELISA tests, patients had known diagnosis.
Wassell 2004[213]	Do not know duration of disease in CDs. The IBS had been diagnosed in previous year - but one year post-diagnosis in IBS group could mean that they had been treated so FC might have been higher at onset.
Wassell 2012[89]	No details of patients providing samples for FC testing
Xiang 2008[214]	Patients already had known diagnosis of CD

Appendix 3. Description of different tests

Enzyme-linked immunoassay (ELISA)

Phical ELISA Test

This is a quantitative ELISA kit manufactured by Calpro (Oslo, Norway). The kit can be used to measure increased concentration of calprotectin in plasma, cerebrospinal fluid, synovial fluid, urine and stool. This test has FDA approval and is marketed in the US for determination of calprotectin level in stools. It has a CE mark.

A polyclonal rabbit antibody is used. The manufacturer states that the affinity of the antibody to six different epitopes of calprotectin makes this ELISA test more robust and less likely to give false results compared to the test using monoclonal antibody, which has affinity for single epitope.

The cut-off value is 50 mg/kg and faecal calprotectin (FC) level above 50 mg/kg is regarded as positive. Previously the cut-off level was 10 mg/l.
[http://www.phical.com/uploads/PhiCaltestperformance.pdf]
[http://www.phical.com/uploads/Instructions.pdf[

PhiCal Calprotectin ELISA kit K6927

This is a quantitative ELISA test manufactured by Immundiagnostik AG (Bensheim, Germany). The kit is supplied by Biohit in the UK. Indications for using this kit include: marker of acute inflammation, estimation of degree of gastrointestinal inflammation, for monitoring Morbus CD, Colitis ulcerosa or the patient status after removal of polyps and discrimination between patients with IBD and IBS.
An older version is K6937. Both versions have CE marks.

The assay uses two monoclonal antibodies that bind to human calprotectin. The sample can be stored but the manufacturer advises against storing samples for >48 hours at 2 to 8˚C. Stool samples can be stored for longer periods at -20˚C.

A limitation of the test, not relevant to this review, is that stool samples with calprotectin level higher than the upper standard value need to be diluted and re-assayed. A faecal calprotectin level above 50 mg/kg is regarded as positive, for adults and children aged 4 to 17 years. The manufacturer however recommends the laboratory to establish their own normal range.

[http://www.immundiagnostik.com/fileadmin/pdf/PhiCalCalpro_Stuhl_1h_K6927.pdf]

EK-CAL

This ELISA kit is manufactured by *Bühlmann Laboratorie (Switzerland)*. It is used for extraction and quantitative determination of faecal calprotectin levels. It has a CE mark.
A monoclonal antibody is used.

The assay can be performed in two different ways, based on the expected faecal calprotectin levels. The low range ELISA procedure can be used for FC levels up to 600µg/g (range 10-600µg/g) and the extended range ELISA procedure for FC levels up to 1800µg/g (range 30-1800µg/g).

The cut-off level is 50µg/g for both adults and children aged between 4 and 17 years.
[http://www.buhlmannlabs.ch/files/documents/core/Inflammation/ifu/ek-cal-ifu-ce-121120.pdf]

Calprest

This is an ELISA kit developed by Eurospital Spa (Trieste, Italy).
CE mark – not mentioned in NICE scoping documents.
The cut-off level is 50 mg/kg. The manufacturer suggests retesting after a short period of time in patients with FC levels between 50 and 100 mg/kg. The FC level above 50 mg/kg is considered as positive.

[http://www.calprotectintest.com/english/calprest.html]

CALPRO Calprotectin ELISA test (ALP)

This quantitative method was developed by CALPRO AS (Lysaker, Norway).

Based on two studies (Johne et al., 2001; Roseth et al., 1992), calprotectin values of <50 mg/kg, >50 mg/kg, 350 mg/kg and 200-40,000 mg/kg represented normal value, positive value, median value in patients with symptomatic colorectal cancers and active, symptomatic IBD respectively.

One limitation of the test is that repeated freeze-thaw cycles of the specimen may affect the accuracy of the test results. The manufacturer cautionsagainst a diagnosis based on a single stool test.

[http://www.calpro.no/products/calprotectin-elisa-test-alp]

Rapid test

Quantum Blue

This is a rapid test manufactured by Buhlmann Laboratories. There are two types of Buhlmann rapid tests i) The lower range Quantum Blue LF CAL (30 to 300 µg/g) and ii) The high range Quantum Blue LF-CHR (100 to 1800 µg/g). The LF-CAL is designed for distinguishing between organic bowel disease and non-organic, or to exclude IBD. The cut-off value of this test is 50 µg/g. The LF-CHR test is follow-up of IBD patients during their therapy.

There are two parts – the test cartridge (to load the stool sample) and the reader. The reader is used to read quantitative concentration of faecal calprotectin. The results are available within 12 to 15 minutes in µg/g calprotectin.
The manufacturer recommends re-testing samples if results are between 30 and 70 µg/g. This zone is regarded as 'grey zone' and corresponds to the 2.5^{th} -97.5^{th} percentile of imprecision around the cut-off of 50 µg/g.

[http://www.buhlmannlabs.ch/core/quantum-blue/]

Prevent ID CalDetect
This is a semiquantitative immunochromatographic rapid test manufactured by Preventis, GmbH (Bensheim, Germany).
The result is interpreted in about 10 minutes.

If there is a solid red control (C) line, then it indicates the test has run correctly. The next test bands (T1, T2, T3) will depend on the concentration of calprotectin. If C, T1 and T2 are visible, then it indicates a calprotectin concentration between 15µg/g and 60 µg/g. If all the test bands (C,T1,T2 and T3) are visible, the it indicates a calprotectin concentration >60 µg/g.

If the control band (C) remains blue or only a test band (T) is visible, then the test is invalid.

[http://www.preventis-online.de/fileadmin/pdf/checksEngl/CalDetect_engl.pdf]

Prevista
This is a chromatographic immunoassay manufactured by GmbH & Co KG (Munich, Germany). The test device has two lines - a test and a control. The test line contains anti-calprotectin antibodies while the control line contains anti-immunoglobulin antibodies, both dried on the membrane.

The results are read within the next 5 minutes. [Details taken from Damms 2008 – no webpage found]

EliA platform (details based on correspondence with manufacturer)

EliA platform is a fully automated calprotectin stool test, manufactured by the Immuno Diagnostics Division of Thermo Fisher (TF IDD) [previously manufactured by Phadia but the company was acquired by Thermo Fisher in 2011].

No details, such as CE mark, were available from the NICE scoping documents

The test was formally launched in November 2012, and is being currently used across 7 sites in the UK. The test is a fully quantitative test which gives results in mg/kg. Four different types of instruments are available namely Phadia 100, 250, 2500 and 5000. They all vary in size and capacity and are designed to meet the requirement of different laboratories. The most commonly used platform in the UK are Phadia 250 and Phadia 100. The test is run as a single test and does not need to be repeated, an advantage over other ELISA tests. The platform is fully automated. The Phadia solution can be added to the existing Phadia systems without the need for further readers and plate washers. The fully automated system reduces laboratory technician workload, time and cost.

Appendix 4. Quality assessment tables

Table 69. Quality assessment of all the included studies

Study	Representative spectrum?	Acceptable reference standard?	Acceptable delay between tests?	Whole sample verified using reference standard?	Same reference standard used?	Were the patients newly diagnosed?	Reference standard results blinded?	Index test results blinded?	Same clinical data as used in test results available in practice?	Uninterpretable / intermediate test results reported?	Withdrawals explained?
Ashorn 2009	Yes	Yes	Unclear	Yes	Yes	Yes	Unclear	Unclear	Yes	Yes	Yes
Bharathi 2005	Yes	Yes	Unclear	No	Yes	Yes	Unclear	Unclear	Unclear	Yes	Yes
[redacted]											
Burri 2012	Yes	Yes	Unclear	Unclear	No	Yes	Yes	Yes	Yes	Yes	Yes
Canani 2006	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Carroccio 2003	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Damms 2008	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Yes	Yes	Yes	Yes
Diamanti 2010	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Dolwani 2004	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Yes	Yes
El-Badry 2010	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Unclear	Yes	Yes
Fagerberg 2005	Yes	Yes	Yes	Unclear	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Garcia 2006	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Unclear	Yes	Yes	Unclear
Henderson 2012	Yes	Yes	Unclear	No	Unclear	Yes	Unclear	Unclear	Yes	Yes	Yes
Kok 2012	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
[redacted]											
Li 2006	Unclear	Yes	Yes	Yes	Unclear	Unclear	Unclear	Unclear	Yes	Yes	Unclear
Licata 2012	Yes	Yes	Unclear	Yes	Yes	Unclear	Yes	Yes	Yes	Yes	Yes
Limburg 2000	Unclear	Yes	Unclear	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Manz 2012	Yes	Yes	Unclear	Yes	No	Yes	Yes	No	Yes	Yes	Yes
Otten 2008	Yes	Yes	Yes	No	Unclear	Yes	Yes	Yes	Yes	Yes	Yes
Schoepfer 2008	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Schroder 2007	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes	Yes
Shitrit 2007	Yes	Yes	Yes	Yes	Yes	Unclear	Yes	Unclear	Yes	Yes	Yes
Sidler 2008	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Yes	Yes	Yes	Yes
Tibble 2002	Yes	Yes	Yes	Yes	Yes	Yes	Unclear	Unclear	Yes	Yes	Yes
Tomas 2007	Unclear	Yes	Unclear	Yes	Unclear	Yes	Unclear	Unclear	Yes	Unclear	Yes
Turvill 2012	Yes	Unclear	Unclear	No	Yes	Yes	Unclear	Unclear	Yes	Unclear	Unclear
Van de Vijver 2012	Yes	Yes	Yes	No	Yes	Yes	Yes	Yes	Yes	Yes	Yes

217

218

Appendix 5. ROC plots generated in RevMan

Figure 27.IBD vs IBS

Figure 28. Organic vs IBS

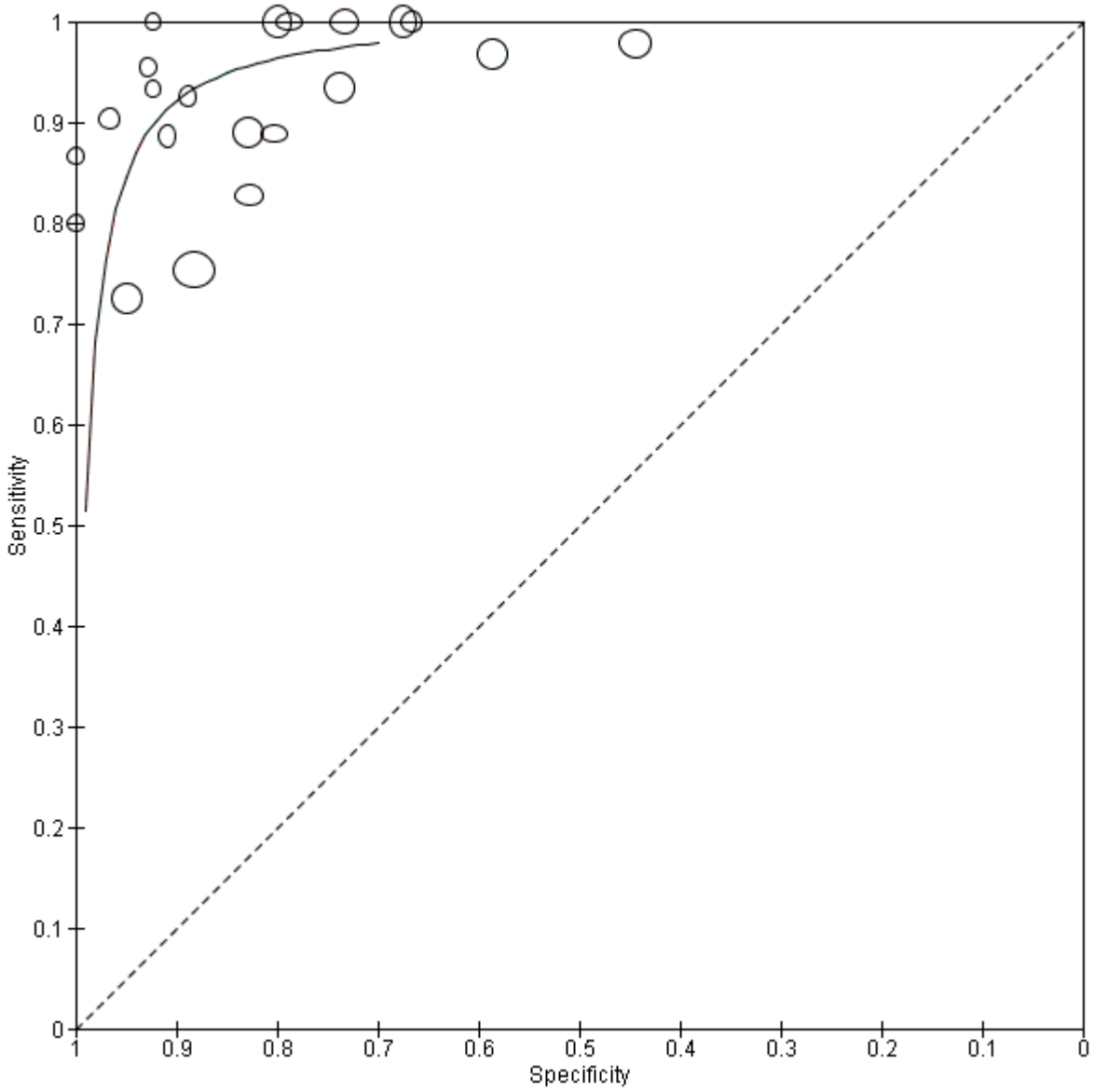


Figure 29. IBD vs non-IBD

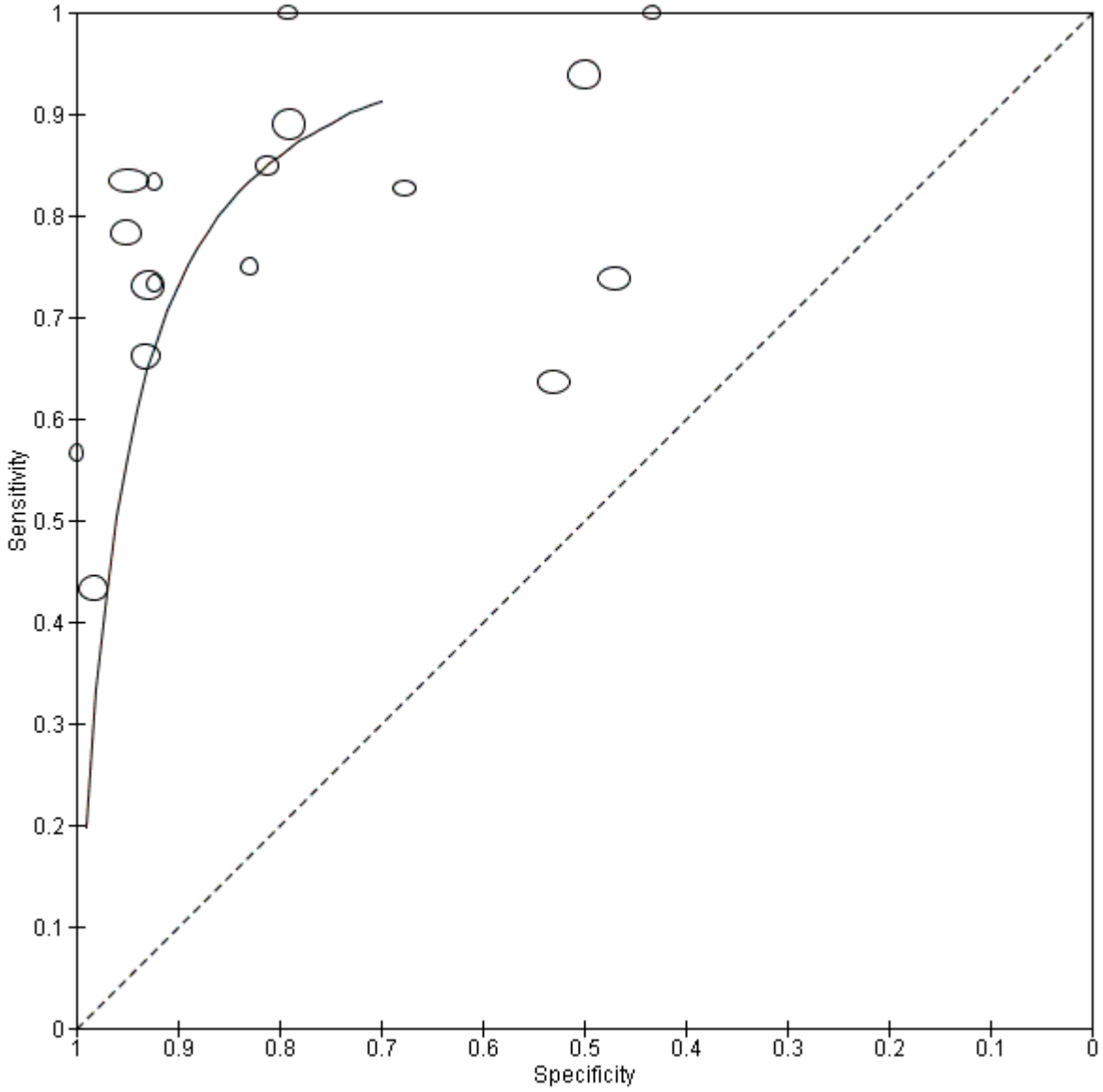


Figure 30. Organic vs non-organic

Appendix 6. Baseline characteristics of all the included studies

Table 70. Baseline characteristics – Table A

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
Ashorn 2009 [79]	To examine the association of FC with serological markers in children and adolescents with IBD	Hospital for children and adolescents in Helsinki, Finland. May 2005-November 2006	55 IBD (incl. indeterminate colitis)=44; non-IBD=11	Median (range) 13.8 (2.7 to 19.9)	Case series	34/36
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]

Bharathi 2005 [meeting abstract][77]	To assess the negative predictive value of FC in excluding bowel pathology in young patients with suspect IBS	Not reported	58
			IBS=42;
			non-intestinal pathology=2;
			miscellaneous=2;
			no significant pathology=12
				Not reported	Case series	Not reported

Burri 2012[86]	To compare three different assays in their ability to identify patients with organic intestinal disease	Department of Gastroenterology of the University Hospital Basel in Switzerland. July 2005-August 2006.	405 Organic intestinal disease: Significant findings=143; No significant findings=262	Median (range) 63 (18 to 97)	A post-hoc analysis of a prospective case series study (Manz 2012)	179/226

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
Canani 2006[60]	To prospectively evaluate the diagnostic accuracy of FC, ASCA/pANCA, IP, and BWUS, independently and in combination.	Pediatric Gastroenterology Unit, Naples, Italy. January-December 2003	45 IBD [CD=17; UC=10]; non-IBD=18 [functional disorders=8, food allergy-related intestinal diseases=5 (celiac disease=2, eosinophilic enterocolitis=2, acute intermittent IgE-mediated disease=1), infectious enterocolitis=4 , and Mediterranean familial fever=1]	Median (IQR) CD group=14.5 (5.1) UC group=11.0 (5.0) Non-IBD group=11.0 (3.3)	Prospective case series	CD=6/11 UC=6/4 Non-IBD=12/6
Carroccio 2003[78]	To evaluate the positive and negative predictive values of the FC assay in identifying the organic causes of chronic diarrhea.	Outpatient clinics of the Division of Internal Medicine of the University Hospital and of the Pediatric Division of "Di Cristina" Hospital, both in Palermo,Italy. January-June 2001	120 (70 adults; 50 children) Children: IBS= 15; organic diarrhea=35; cow's milk intolerance=15; Celiac disease=13; multiple food intolerances=5; intestinal giardiasis=2 Adults: IBS=40; organic diarrhea=30; Celiac disease=10; multiple food intolerances=2; colorectal cancer/adenomatous polyps=3; microscopic colitis=2; diverticulosis/diverticulitis=4; CD=9	Median (range) Adult group=35 (18-72); Paediatric group=3.5 (8 months to 10 years)	Prospective case series	Adults=30/40 Children=20/30

Damms 2008[53]	To evaluate the diagnostic accuracy of the new calprotectin rapid test compared to an established ELISA test	Gastroenterological departments of 3 hospitals and 3 outpatient gastroenterologies, based in Stuttgart, Germany.	84
			Diverticulosis=18;
			adenoma=29;
			carcinoma=8;
			active IBD=18;
			intestinal infections=11
				Mean (range)
				58 (20 to 85)
					Open multicenter case control study	62/78

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
Diamanti 2010[80]	To assess the diagnostic precision of a FC assay, compared to histology, as a stool-screening biomarker for IBD	Gastroenterology and Nutrition Unit of 'Bambino Gesu' Children's Hospital, Rome January 1999-December 2007	197 IBS (normal mucosa)=28 aspecific colitis and benign lymphoid hyperplasia=52 IBD =117 (UC=58; CD=49))	Median (range); Group B (normal mucosa)=9 (2 to 18); Group C (aspecific colitis and benign lymphoid hyperplasia)=11 (1 to 18); Group D (IBD)=12 (1 to 18)	Prospective case series	Group B=15/13; Group C=29/23; Group D=65/52

Dolwani 2004[42]	To compare the utility of a single FC estimation to barium follow through (BaFT) in exclusion of intestinal inflammation	Majority of cases were recruited from a gastroenterology outpatient clinic with a special interest in IBD at the University of Wales Hospital.	63
			Small intestinal Crohn's=6,
			jejunal lipoma=1,
			Roux loop ulceration=1,
			caecal carcinoma=1;
			Crohn's colitis=5;
			coeliac disease=1;
			normal BaFT=48
				Median (range)
				47 (17 to 86)
					Comparative case series	20/43

El Badry 2010[76]	To evaluate the sensitivity and diagnostic accuracy of FC assay at different cut-off values in discriminating between functional and organic GI disorders	Internal Medicine Department in Cairo University Hospital November 2008-August 2009.	29 IBS=20 IBD=9 [UC=8; CD=1]	Mean (SD) IBS group=39.4 (15.9) IBD group=34.3 (15.75)	Prospective case series	IBS=9/11 IBD=6/3

Fagerberg 2005[81]	To determine if FC can be used as a diagnostic test of colonic inflammation to identify those children who require colonoscopy.	Department of Gastroenterology, Astrid Lindgren Children s Hospital, Stockholm, and the Department of Pediatrics, Central	36
			IBD= 22 [CD=10; UC=7; indeterminate colitis=3; juvenile colonic polyposis=1; unspecfied proctitis=1];
			functional bowel disorder=5;
			food intolerant=4;
			spirochetosis=1,
				Median (range)
				Inflammed group= 13.6 (6.7 to 17.8)
				Non-inflammed group=14.2 (6.5 to 17.3)
					Prospective comparative case series	Inflammed=11/11
						Non-inflammed=6/8

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
		Hospital, Vasteras, Sweden.	one teenager had nutritional iron deficiency anemia=1, improved spontaneously=3			

Garcia 2006 (translated from Spanish)[54]	To assess the usefulness of FC to predict the presence of pathological colonoscopy	Hospital Universitario Reina Sofia Córdoba, Spain	190
			normal colonoscopy=117 [of which IBS=9];
			colon adenoma (polyps)=28;
			colorectal cancer (CRC)=20;
			IBD=25 [UC=9; CD=16]
				Mean
				Normal colonoscopy=59.5;
				Colon adenoma (polyps)=60.2;
				CRC=71.8;
				IBD=35.8
					Prospective case series	Normal colonoscopy=69/48
						Colon adenoma (polyps)=9/19
						Colorectal cancer (CRC)=13/17
						IBD=11/14

Henderson 2012[26]	To describe the differences in FC levels between IBD types (CD, UC,and IBD-U) and non-IBD disease categories.	The pediatric gastroenterology department Royal Hospital for Sick Children in Edinburgh. January 1 2005-December 31 2010.	190 IBD group=91 [CD=62; UC=21, IBD-U=8] Control (non-IBD) group=99 [IBS=32]	IBD group=12.6 (9.5 to 14.0) Control group=9.3 (5.2 to 12.7)	Retrospective case control	IBD group=56/35; Non-IBD control group=55/44
Kok 2012[33]	To quantify the diagnostic accuracy of 3 biomarker tests for the inclusion or exclusion of OBD in patients with persistent lower-abdomen complaints	Primary care. Data from the CEDAR study in 170 general practices in 2 regions of the Netherlands July 2009-January 2011	382 Organic bowel disease (OBD)=99 [adenoma=53.5% of OBD (adenoma > 1 cm=30% of adenomas; ≤1 cm=70% of adenomas); IBD=19 (19%) of OBD]; Non OBD=283	Median (range) 60 (18 to 91)	Data from the CEDAR study, an ongoing, prospective, cross-sectional, diagnostic study	175/211

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Li 2006 [translated from Chinese][55]	To assess the value of FC in differential diagnosis of IBS	Both outpatients and hospitalized patients of Peking University Third Hospital 2004-2005.	240 IBS=60; Chronic inflammation group=60 [UC=33; CD=15]; colorectal cancer group=60; healthy controls=60	Mean (range) IBS=51 (18) Chronic inflammation=42 (16)	Prospective comparative study	IBS=30/30; Chronic inflammation=26/34
Licata 2012[56]	To assess the diagnostic performance of FC as a stool-screening biomarker for organic intestinal disease	Gastroenterology outpatient department (Gastroenterology and Hepatology Unit, University of Palermo) March 2004-May 2009	346 No inflammation=204 [IBS=197; diverticular disease=7]; Inflammation=142 [IBD=82 (CD=56; UC=26); microscopic colitis=6; diverticular disease= 4; polyps=6; ischemic colitis=1; nonspecific colitis=22; IBS=21]	Median (range) No inflammation= 38 (18 to 87) Inflammation= 41 (17 to 80)	Prospective case series	No Inflammation= 74/130; Inflammation= 71/71
Limburg 2000[70]	To assess and compare calprotectin and haemoglobin as stool screening biomarkers for colorectal inflammation	The Mayo Clinic (Rochester, MN) November 1996-July 1998	110 Inflammation=29 [Crohn's or UC=16; microscopic or collagenous colitis=11; other inflammatory conditions=2]; No inflammation=81 [histologically-confirmed normal mucosa=49; macroscopically normal mucosa (without biopsy)=11; polyps (all , 1 cm in diameter) but otherwise normal-appearing mucosa=21]	Mean (SD)=57 (16)	Prospective case series	With colorectal inflammation= 10/19; Without colorectal inflammation= 30/51

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
Manz 2012[88]	To prospectively investigate the value of FC as a biological marker for the diagnosis of intestinal organic disease in symptomatic patients	Department of Gastroenterology of the University Hospital Basel in Switzerland.	538 No clinically significant finding=326 [normal finding=314; hyperplastic polyps=12]; Clinically significant finding=212 [esophagitis=31; erosive gastritis/duodenitis=22; gastric ulcers=11; gastric carcinomas=3; colitis/ielitis=53 (infectious colitis=8; CD=10; UC=16; diverticulitis=13; microscopic colitis=5; ischemic colitis=1); adenomatous polyps=50; colorectal cancers=17]	Mean (IQR) Referred for colonscopy=63 (53 to 71) Referred for EGD=55 (42 to 65)	Prospective case series	Referred for colonoscopy=173/218 Referred for EGD=75/147
Otten 2008[65]	To evaluate the diagnostic accuracy of two new rapid calprotectin and lactoferrin faecal tests in assessing colonic inflammation	Gelderse Vallei Hospital, The Netherlands. May-June 2007	114 IBS=91; IBD=23 [CD=6; UC=5; unspecified colitis=12]	Mean IBS=52.3 IBD=44.5	Cross-sectional design	IBS=42/49 IBD=11/12
Schoepfer 2008[67]	To measure the accuracy of faecal markers, CRP, blood leukocytes, and IBD antibodies for discriminating IBD from IBS	Outpatients and inpatients from the Departments of Gastroenterology of the University Hospital Bern and Kantonsspital Lucerne. April 2005-October 2006.	94 IBD=64 [CD=36; UC=28]; IBS=30	Mean (SD) [range] CD=41 (18) [20 to 78] UC=45 (14) [23 to 72] IBS=40 (19) [20 to 79]	Prospective case series	Overall IBD=32/64 CD=17/19 UC=15/13 IBS=8/22
Schroder 2007[69]	To evaluate calprotectin, lactoferrin and PMN-elastase in faeces to detect active GI inflammation	J.W. Goethe-University, Frankfurt am Main, Germany (1st Department of Internal Medicine) August 2002 and January 2004	76 IBD=45 [CD=25; UC=20]; IBS=31	Median (range); CD=40 (25 to 59) UC=38 (24 to 75) IBS=43 (20 to 72)	Prospective case series	Total=33/43 CD=7/18 UC=15/5 IBS=11/20

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
Shitrit 2007[57]	To assess the predictive value of FC in organic colonic disease.	Department of Gastoenterology, Shaare-Zedek Medical Center, Israel	69 Abnormal histology=28 [IBD=11 (CD=7); carcinoma=12; polyps=5]; Normal histology=41	Not reported	Case series	Not given
Sidler 2008[82]	To define the appropriate roles for faecal S100A12 and calprotectin in the initial investigations of children with suspected IBD	Gastroenterology Outpatient Clinic at Sydney Children's Hospital, Randwick, Australia	61 IBD=31 [CD=30; UC=1]; Non-IBD=30	Mean (SD) [range] IBD=11.9 (3.3) [2.4 to 16] Non-IBD=10.3 (3.6) [2.2 to 15.5]	Prospective case series	IBD=19/12 Non-IBD=17/13
Tibble 2002[43]	To determine if the use of FC and intestinal permeability are useful in differentiating between patients with organic and nonorganic disease.	Gastroenterology outpatient department of a teaching hospital in South London.	602 Organic disease=263 [small bowel (CD=84; Celiac disease=12; infective diarrhea=9; small bowel enteropathy=21; diabetic diarrhea=50); colonic (CD=18; UC=87; microscopic colitis=5; collagenous colitis=1; diverticular disease=14; cancer=7)] Nonorganic disease=339 [IBS=275; IBS + nonulcer dyspepsia=38; IBS + other=26]	Median (range) 40 (18 to 90)	Prospective case series	231/371
Tomas 2007[61]	To evaluate FC in paediatric patients with signs and symptoms suggestive of IBD	Patients with gastrointestinal symptoms who had been referred to the Pediatric Gastroenterology Unit of Son Dureta University Hospital (Palma de Mallorca), Spain. 2003-2005	43 Functional pathology=13; Organic disease=30 [IBD=15]	Mean (range) 10.1 (3 months to 15.3 years)	Retrospective case series	Not reported
Turvill 2012[40]	To determine the NPV of a normal FC in excluding	The Department of Gastroenterology,	630 Cohort 1 (normal FC)=500 ;	Mean (range) 41 (16 to 60)	Retrospective cohort study	Cohort 1 (FC<50):

Study ID	Aim of study	Setting & Dates	Number of participants	Age (years)	Study Design	Gender (M/F)
	organic intestinal disease in patients with intestinal symptoms	York Hospital. January 2004-May 2007	Cohort 2 (raised FC)=130 Organic disease=109; Nonorganic disease=521		from patient records.	145/355 Cohort 2 (FC >50): 43/87
Van de Vijver 2012[74]	To determine a diagnostic strategy to minimise the number of patients with negative endoscopy results without missing any cases of IBD	Paediatric outpatient clinics of six general hospitals and one tertiary care hospital in the northern region of the Netherlands. February 2009-June 2010	117 IBD=42 [CD=24; UC=16; IBD unclassified=2]; Non-IBD=75	Mean (range) 14 (6 to 18)	Prospective diagnostic accuracy study	Confirmed IBD=19/26 Non-IBD=38/37

Table 71. Baseline characteristics-Table B

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Ashorn 2009 [79]	Suspicion of IBD	Not reported	Phical Test, Calpro AS, Oslo, produced by NovaTec Immunodiagnostica, Dietzenbach, GmBH, Germany - ELISA ≥100 µg/g	Not reported	Upper gastrointestinal biopsies using the modified Sydney system. All underwent upper and lower endoscopies. Diagnosis of IBD made on histopathological criteria.	Paediatric Research Foundation; No disclosures
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Bharathi 2005 [meeting abstract][77]	Presented with abdominal pain and/or loose stools	Not reported	PhiCal - ELISA 60 µg/g	Not reported	In 38/42 patients the diagnosis of IBS made after appropriate, targeted investigations (OGD, flexible sigmoidoscopy, colonoscopy, ultrasound, small bowel studies, blood tests).	No disclosures. No funding sources mentioned.
Burri 2012[86]	Patients undergoing endoscopy of the gastrointestinal tract for abdominal discomfort at the Department of Gastroenterology of the University Hospital Basel in Switzerland	Patients younger than 18 years old were excluded	Calprotectin (EK-CAL, Bühlmann Laboratories, Switzerland)- ELISA PhiCal, Calpro AS, Norway) -ELISA both used cut-off levels ≥ 50 µg/g [manufacturer's recommended cut-off]	Samples collected at home 24 hours prior to bowel preparation for endoscopy-delivered on day investigation. Stored in a refrigerator - transferred to lab within 48 hours for analysis.	Final diagnosis independently adjudicated by two gastroenterologists on the basis of all the patient's available medical records (clinical data, laboratory values, endoscopy report, histology report). 70 patients (17.3%) also received esophagogastroduodenoscopy	Independent funding. Buhlmann Laboratory AG provided the assays. All authors declared no conflict of interest.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Canani 2006[60]	All children referred to the Paediatric Gastroenterology Unit for initial assessment of suspected IBD.	Patients with symptoms or signs (right-lower quadrant mass or perianal disease or hematochezia) mandating a complete work-up for IBD.	Calprest, Eurospital Spa, Trieste, Italy - ELISA 95.3 mg/g	Stool samples were collected before endoscopy, stored at -20˚C and thawed at room temperature before testing.	Three expert paediatric gastroenterologists made a final diagnosis of IBD or non-IBD according to the presence or absence of previously reported clinical, radiographic, endoscopic and histopathologic criteria. Bowel wall ultrasonography performed in all patients within 24 hours of admission.	No disclosures. No funding sources mentioned.
Carroccio 2003[78]	A history of chronic diarrhea of unknown origin, lasting for more than 4 weeks, with or without abdominal pain.	Previous evaluation for chronic diarrhea; overt GI bleeding, sigmoidoscopy or colonoscopy during the previous 2 years for any cause; familial adenomatous polyposis and hereditary onpolyposis; colorectal cancer	Calprest; Eurospital - ELISA 50 µg/g and 100 µg/g [<50 =negative; 50–100=borderline; >100 =positive]	One stool sample collected and returned within 1 week of first visit. Samples stored at -20˚C. Two aliquots from a single stool sample from each participant were assayed within 4 weeks, and the mean of the two measurements recorded.	All patients evaluated by the Rome criteria for IBS and haematology and chemistry tests. Adults also had biopsy with sigmoidoscopy or colonoscopy. If positive at the first-step, then underwent a diagnostic work-up. Children with positive occult blood in the stool or with serum indices of inflammation had colonoscopy with biopsy.	No disclosures. No funding sources mentioned.
Damms 2008[53]	At least 18 years old and underwent colonoscopy according to a medical indication (GI disorders) or for CRC screening–preventive medical checkup.	Known extraintestinal inflammatory diseases. Patients whose medical history included NSAID drugs or anticoagulants.	Bühlmann Laboratories AG (Schönenbuch, Switzerland) - ELISA Prevista GmbH & Co KG, Munich, Germany) - semiquantitative rapid test 50 µg/g [manufacturer's recommended cut-off]	A cold chain was maintained for all samples throughout. Specimens preserved at −20°C, and assayed within the next 3 months. Patients provided a single stool before colonoscopy.	Patients underwent colonoscopy according to a medical indication (gastrointestinal disorders) or for CRC screening-preventive medical check-up.	No disclosures. No funding sources mentioned. Support for analytical kits from Prevista GmbH, Munich

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Diamanti 2010[80]	Recurrent abdominal pain and altered bowel habit, associated with one or more symptoms including rectal bleeding; loss of weight; abnormality of physical examination; delay of pubertal development; positive clinical history; altered blood tests.	Not given	Calprest, Eurospital, Trieste, Italy 100 µg/g and 160 µg/g	Two stool samples were collected before purgation and an average of the two calprotectin measurements was recorded. Faecal calprotectin was measured on frozen (-20˚) stool specimens	Colonoscopies including intubation of terminal ileum. Mucosal biopsy samples were taken from the terminal ileum, the cecum, the ascending colon, the transverse colon, the descending colon, and the rectum. An experienced paediatric histopathologist, assessed each biopsy specimen.	No disclosures. No funding sources mentioned.
Dolwani 2004[42]	Consecutive patients undergoing small bowel BaFT examination as part of their clinical workup after presenting with abdominal pain and or diarrhoea	Patients with: known malignancy, on NSAIDs, coeliac diseases, evere cardiopulmonary, renal or hepatic impairment, significant	ELISA-based method (supplied by Calprotech Ltd, London, UK) - brand not specified 60 µg/g [determined by comparing results and using co-ordinates of ROC curves]	One faecal sample within 7 days before, or 7-10 days after BaFT. A portion of stool frozen at -20˚C and then samples were assayed into batches. On days 0, 1, 3 and 7 a sample of stool was taken and frozen for later analysis.	Patients undergoing barium follow through (BaFT) also underwent rigid sigmoidoscopy and stool cultures as part of their workup and those with abnormal rigid sigmoidscopy or positive stool cultures were excluded.	Funded by grant from the Wales office of Research Department No disclosures.
El Badry 2010[76]	Presence of symptoms for at least six months suggestive of organic pathology, including intense abdominal pain, chronic diarrhoea, weight loss and/or anorectal bleeding. Also, to have had an endoscopic and/or intestinal radiological procedure at the initial hospital visit	Regular intake of NSAIDs, aspirin, and/or anti-coagulants, or the concomitant presence of other non-GI diseases, e.g. rheumatoid arthritis, other connective tissue inflammatory diseases or liver cirrhosis.	PhiCal, produced by Nycomed Pharma AS - ELISA >50 µg/g and > 100 µg/g [Normal reference value was <15µg/g and the manufacturer established the margin values between 50 and 100 µg/g].	A single stool sample was collected from each patient and stored in a suitable container at -20^{o} C until assayed for calprotectin. The faecal sample was delivered 3 days before colonoscopy.	All patients evaluated using ROME III criteria for IBS. Also, had full medical history with thorough clinical examination; stool analysis and culture to exclude infections; complete blood picture, abdominal ultrasound examination; and complete colonoscopy with intubation of the terminal ileum including multiple biopsies from the lesions for histopathological evaluation.	No disclosures. No funding sources mentioned.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Fagerberg 2005[81]	Children with gastrointestinal symptoms who were scheduled for colonoscopy to rule out IBD.	Had no bacterial gastroenteritis detectable by faecal culture or serology and did not have any other chronic inflammatory disease.	Calprest , Eurospital SpA, Trieste, Italy - ELISA ≥50 µg/g	The stool samples were prepared and analysed for calprotectin according to the manufacturer s instructions. Stool was sent the same day or next day by mail to the laboratory. After extraction, the supernatant was collected and frozen at -20˚C.	Decision to perform a colonoscopy based on the child's medical history, physical examination and blood tests. In general, complete ileocolonoscopies were performed. Histopathologic evaluations of colorectal inflammation were made for diagnosis of IBD. All children with non-inflamed colonic mucosa had a complete colonoscopy, as did most of the children with inflammation.	County Council of Bvastmanland, Swedish Society of Medicine, Mayflower Charity Foundation for Children, Sweden No disclosures.
Garcia 2006 (translated from Spanish)[54]	Consecutive individuals who underwent colonoscopy for medical indications	Patients with severe cardiopulmonary disease, kidney or liver disease, celiac disease, known malignancy and patients with other organic processes in different colonoscopy polyps, colorectal cancer or IBD	Calprest, Eurospital, Trieste, Italy - ELISA 217 mg/kg	All patients collected a stool sample one day before the colon preparation for the determination of the FC.	All patients had a complete colonoscopy. The diagnosis of IBD was based on clinical criteria, endoscopic and histologic findings.	Funded by Eurospital and Schering Plough No disclosures.
Henderson 2012[26]	All patients potentially undergoing endoscopy before 18 years of age in South-East Scotland. IBD group: All incident cases of PIBD diagnosed by standard criteria and had FC measured . Ccontrol patients identified from	Aged < 1 year or >18 years of age on the endoscopy date; greater than a 6-month delay between the FC sample and the endoscopy date; FC sample taken after endoscopy; any previously known, hospital diagnosed, GI disease; and previous	PhiCal Test (Calpro AS, Lysaker, Norway) >50, 100, 200, 300, and 800 µg/g [<50 µg/g: normal; 51-100 µg/g: possible gastrointestinal (GI) inflammation;101-200 µg/g: GI inflammation; >200 µg/g: active GI	FC measured according to manufacturer's instructions	IBD patients: all incident cases diagnosed by standard clinical, histological and radiological findings. Non-IBD (control) patients: all had undergone both upper and lower endoscopy for the clinical suspicion of paediatric IBD.	Medical Research Council project grant; Chief Scientist Office in Scotland and Cure Crohn's Colitis. No conflict of interest.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
	hospital records and departmental endoscopy lists.	upper or lower GI endoscopy.	inflammation]			
Kok 2012[33]	Patients consulting their GPs for persistent lower-abdominal complaints; high risk of OBD (as lower-abdomen complaints for > 2 weeks plus 1 or more of the following, rectal bleeding, altered defecation pattern, abdominal pain, fever, diarrhea, weight loss, sudden onset in the elderly, physical findings suggestive of OBD	<18 years old, unable to give informed consent, previously diagnosed with OBD, or positive on triple faeces test, not requiring endoscopy.	Quantum Blue Buhlmann Laboratories - POCT EK-CAL (Buhlmann) - ELISA >50 µg/g [manufacturer's recommended cut-off]	Faecal sample collected directly following inclusion into the study and kept refrigerated at all times. Samples processed directly in 38% of cases and initially frozen in 62%.	OBD determined at endoscopy as performed by experienced gastroenterologists, taking biopsies if required according to routine clinical practice. All patients with an inconclusive diagnostic reference procedure followed for 3 months to establish a definite diagnosis. Colonoscopy performed in 91.9% of patients, sigmoidoscopy in 5.5%, and other bowel examinations in 10 2.6% . 89.9% of .OBD was confirmed by histology	Funded by Netherlands Organization for Health R&D. Alere Health, provided iFOB POC tests. Buhlmann provided the calprotectin POC and ELISA test. No conflict of interest.
[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Li 2006 [translated from Chinese][55]	IBS group: all confirmed by Rome II criteria; chronic inflammation group - confirmed by colonoscopy or operation or pathologic diagnosis; Control group: people who were cured from a polyp of the intestinal tract by endoscopic therapy	Absence of overt upper GI symptoms or stomach/small intestine disease; severe disease of the heart, lung, liver, kidney, nerve, or mental disorder. Colorectal adenomas excluded.	PhiCal Test(Bio-Rad 550) made by Finland Biohit company - ELISA 50 mg/kg [set according to references as well as the recommended value from the instructions of the assay kit]	Faecal samples collected within one week of endoscopy or before surgical operation, transferred to hospital within 2 hours of collection, sealed and stored under -20℃, in preparation for the FC assay.	The IBS group all confirmed by Rome II criteria, and no abnormality was seen by colonoscopy or colon contrast. The chronic inflammation group were confirmed by colonoscopy/operation/pathologic diagnosis.	No funding sources mentioned. No disclosures.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Licata 2012[56]	Age of at least 18 years; consecutive patients referred to evaluate chronic (≥4 weeks) nonbloody diarrhea of unknown origin.	Overt GI bleeding, known colorectal or gastric neoplasia, familial adenomatous polyposis and hereditary nonpolyposis colorectal cancer syndrome, history of colonic surgery, recent intake of NSAIDs, aspirin or anticoagulants.	Calprest (Eurospital SpA, Trieste, Italy) - ELISA >150 µg/g	Stool samples collected and returned by each participant in a disposable device to avoid toilet water artefacts and to simplify laboratory sampling. Upon receipt, samples were aliquoted for immediate assay or stored at -20˚ until assay.	All patients underwent colonoscopy with biopsies. Histopathologic evaluations of colorectal inflammation were made by experienced GI histopathologists. Histologic inflammation was defined by histologic standard criteria and subtyped as: Crohn's disease, ulcerative colitis, microscopic colitis.	No funding sources mentioned. No disclosures.
Limburg 2000[70]	Adult patients who had been referred for colonoscopy to evaluate chronic diarrhea (≥4 week duration) of unknown origin or chronic colitis of unknown activity.	Abnormalities on GI x-rays, overt GI bleeding, GI endoscopy performed within the preceding 2 wk, known colorectal neoplasia, familial adenomatous polyposis, and hereditary nonpolyposis colorectal cancer syndrome.	PhiCal (Nycomed Pharma AS, Oslo, Norway) - ELISA 100 µg/g [preset threshold values supplied by the manufacturer (<50 mg/g of stool = negative, 50–100 mg/g of stool =weakly positive, and >100 mg/g of stool = strongly positive)]	One stool specimen was collected and returned by each participant using a disposable device to avoid toilet water artifact and simplify laboratory sampling. Upon receipt, samples were aliquoted for immediate essay or stored at -70˚C until assay performance.	Colonoscopies were performed by experienced staff gastroenterologists, unaware of the faecal assay results. Caecal intubation, coupled with ≥90% mucosal surface visualization, constituted a complete examination. Mucosal abnormalities were recorded by anatomic subsite and biopsies were obtained when clinically indicated.	NIH Grant plus a grant from Nycomed Pharma, Oslo, Norway [the manufacturers of the PhiCal ELISA). No disclosures.
Manz 2012[88]	Patients undergoing endoscopy of the GI tract for abdominal discomfort - was defined as any sensation of any quality and intensity of abdominal pain. If several symptoms were present, abdominal discomfort had to be the main symptom.	Patients younger than 18 years old.	EK-CAL (Bühlmann Laboratories AG, Schönenbuch, Switzerland) - ELISA 50 µg/g [manufacturer's recommended cut-off]	A single stool sample was collected from each participant 24 hours prior to bowel preparation for endoscopy. Samples delivered on the day of the investigation and stored in a refrigerator before transfer to the study laboratory within 48 hours of analysis.	All patients underwent standard endoscopies performed by 4 senior gastroenterologists. Biopsies were collected if appropriate as decided by the endoscopist. Patients with no significant lesion but FC levels > 50 µg/g on initial endoscopy were further investigated with either EGD or colonoscopy. The	Researchers were independent of funding. One author is an employee of Buhlmann Laboratories. All authors declared no conflict of interest.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
					endoscopists performing the follow up endoscopy were aware of the reason for the investigation (positive test).	
Otten 2008[65]	Consecutive patients with lower gastro-intestinal abdominal complaints, including bloating, change in defecation frequency or consistency, or blood and mucus in the faeces, referred for endoscopy or sigmoidoscopy to the endoscopy unit.	Younger than 18 years of age, a history of colonic surgery and those with iron deficiency	Prevent ID CalDetect Preventis, Bensheim, Germany) - POC Test 15, 50 and 60 µg/g [in line with the instructions of the manufacturer, the test was evaluated positive when at least the second test line appeared]	On the day of endoscopy patients returned their faecal samples. Faecal rapid tests were performed at the endoscopy unit before the procedure. Faecal samples were stored at -20˚C for a maximum of 1 month for ELISA test at a later stage.	All patients underwent colonoscopy or sigmoidoscopy according to routine procedure. According to routine clinical practice, the diagnosis was based on the endoscopic picture; biopsies were taken if necessary to confirm the diagnosis. In half of the patients with IBD, biopsies were taken.	No disclosures. No funding sources mentioned. Orange Medical supplied the calprotectin tests.
Schoepfer 2008[67]	Age 18–80 years, complete colonoscopy with intubation of terminal ileum including biopsies, faecal samples delivered from 3 to 1 days before colonoscopy and after the evaluation, an established diagnosis of bowel disease.	Incomplete ileocolonoscopy, microscopic colitis, infectious ileocolitis, colorectal cancer, colorectal polyps, history of colorectal or small bowel surgery, regular intake of aspirin and/or an NSAID.	PhiCal Test delivered by CALPRO AS (Oslo, Norway) - ELISA 15, 50 and 60 µg/g [in line with the instructions of the manufacturer, the test was evaluated positive when at least the second test line appeared]	Patients collected faecal samples in three faecal tubes. Samples collected from the outpatients were sent by urgent mail from Monday to Thursday so that no samples arrived on weekends. All samples were processed within 48 hours after collection.	IBD diagnosis based on symptoms , clinical examination, endoscopic findings, histologic analysis, radiologic workup, and laboratory tests. All IBS patients fulfilled the Rome II criteria and had an endoscopy of the upper GI tract. The decision to perform an examination of the jejunum and proximal ileum was left to the judgment of the treating gastroenterologist.	Funded by Swiss National Science Foundation. No disclosures.
Schroder 2007[69]	History of chronic diarrhoea of unknown origin, lasting for more than 4 weeks, with or without abdominal pain	Previous evaluation for chronic diarrhoea, overt gastrointestinal bleeding, sigmoidoscopy or colonscopy during the	Immundiagnostik AG, Bensheim Germany - ELISA 15 µg/g [manufacturer's recommended cut-off]	Fresh stool samples provided for determination within 1 week prior to colonoscopy. Samples received within 24 hours	Colonscopies with biopsies from each segment of the colon. Mucosal abnormalities recorded by anatomic location. Inflammation was defined and	Partly funded by the Else Kroner-Fresenius-Foundation, Germany.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
		previous 2 months peformed for any cause, familial adenomatous polyposis and hereditary non-polyposis colorectal cancer syndrome and pregnancy		of defecation. Upon receipt, stool was aliquoted for immediate assay or stored at -20˚C until assay performance.	graded by standard histological criteria and subtyped by endoscopic and histological features	The authors declared no conflict of interest.
Shitrit 2007[57]	Patients referred to the Department of Gastroenterology for colonoscopic examination of various indications, including screening.	Intake of NSAIDs and/or antibiotics during the three months preceding the study, concomitant serious illness, pregnancy, alcohol abuse, and evidence of a respiratory tract infection	Calprest, Eurospital, Trieste, Italy - ELISA 150 mg/kg [Normal level described as 25 mg/kg]	Stool samples collected before colonoscopic preparation. The samples were stored in a household freezer and they were brought on the day of examination. The samples were frozen at -20˚C until assayed.	Colonoscopy and histopathology.	No disclosures. No funding sources mentioned.
Sidler 2008[82]	Age between 2 and 18 years, and presenting with GI symptoms suggestive of an organic gut disease and required further investigation based on clinical assessment. Symptoms included chronic diarrhea over more than 1 month, bloody stools, and abdominal pain occurring for at least a month.	Children with a previously established diagnosis of an organic gastrointestinal disease; infectious gastroenteritis was excluded by at least 2 negative stool cultures; having used NSAIDs, antibiotics, or corticosteroids in the preceding 2 weeks	PhiCal test, Nycomed, Oslo Norway - ELISA 50 mg/kg	Prior to admission for GI endoscopy and colonscopy, children provided a stool sample collected at home before the bowel preparation in a sterile collection vessel, stored briefly at -20˚C (home freezer), and transported frozen to the laboratory, where samples were stored at -80˚C until analysis.	All children underwent upper GI endoscopy and complete ileocolonoscopy Multiple tissue samples were assessed by an experienced paediatric histopathologist. Final diagnosis based upon standard diagnostic criteria including clinical, endoscopic, histological, and imaging findings.	Partly funded by the Foundation Eugenio Litta, Geneva and Freiwillige Akademische Gesellschaft, Basel, Switzerland. No disclosures.
Tibble 2002[43]	Referred to a gastroenterology outpatient department by GPs. All patients had clinical symptoms suggestive of	Previously known diagnosis of IBD, colorectal carcinoma, and serious cardiopulmonary, hepatic, renal, neurologic,	In-house ELISA -not a commerically available [<10 mg/L - converted to 50 µg/g]	Patients provided a single stool sample for measurement of calprotectin that was submitted within 48 hours.	Patients were classified investigator as having positive or negative Rome I criteria. Each patient underwent one or more invasive diagnostic	Supported by NHS Executive South Thames Regional Office No disclosures.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
	organic intestinal disease or IBS that had not responded to therapy instituted by n primary care and were of sufficient severity for further investigation to exclude organic pathology.	and psychiatric disease, or referred for investigation of symptoms of oesophageal reflux, symptoms associated with gastroesophageal pathology, or dyspepsia.			imaging procedures of the GI tract as the gold standard, appropriate to their symptoms.	
Tomas 2007[61]	New patient referrals from primary care, aged 16–60 years, with intestinal symptoms, defined as any of change of bowel habit, abdominal pain, bloating, mucorrhoea, bleeding, tenesmus or urgency.	Not reported	Calprest, Eurospital, Trieste, Italy 50, 100, 150, 200 µg/g	A single stool sample was collected from each patient in a plastic container, which was sent to the laboratory in less than 48 hours; samples were then frozen at -70˚C until analyzed.	Diagnosis based on clinical criteria, laboratory, image and endoscopic test results, in relation with their evolution, and complied with the functional pathology criteria established in the Rome II meeting.	No disclosures. No funding sources mentioned.
Turvill 2012[40]	New patient referrals from primary care, aged 16–60 years, with intestinal symptoms. These were defined as any of change of bowel habit, abdominal pain, bloating, mucorrhoea, bleeding, tenesmus or urgency.	Patients with fast track colorectal symptoms.	PhiCal - ELISA 50 µg/g [manufacturer's recommended cut-off]	On request of a FC, a stool sample was delivered by the patient either to the hospital or to their primary care provider. It was then forwarded internally to laboratory.	Cohort 1: 43% of patients had a full evaluation of the colon by colonoscopy or barium enema and 60% had supportive histology. 53% had no investigations. Cohort 2: extensive intestinal investigation - inlcuding colonoscopy, barium enema, barium meal, CT enterography, capule endoscopy and supportive histology in 83% of patients.	No funding sources mentioned. The authors declared no conflict of interest.

Study ID	Inclusion Criteria	Exclusion criteria	FC Test and Cut-off Used	Details of FC testing	Reference standard	Funding source or conflict of interest
Van de Vijver 2012[74]	Aged between 6 and 18 years of age with abdominal complaints and with a clinical suspicion of IBD who fulfilled the clinical criteria for IBD.	Younger children (who have higher normal values of faecal calprotectin).	Calpro, Calpro AS, Lysaker, Norway - ELISA 50 µg/g [manufacturer's recommended cut-off]	After the first presentation at the outpatient clinic, all patients provided a stool sample collected at home.	Paediatricians, referred 68 of 117 patients for endoscopy on the basis of a high index of suspicion for IBD. Majority did not need endoscopy to exclude IBD, and had other tests, including stool analyses for bacteria, ova and parasites, gastroscopy, abdominal ultrasound, CT scan, Meckel scan, serology and dietary measurements leading to the diagnosis.	No funding sources mentioned. The authors declared no conflict of interest.

Appendix 7. Cost effectiveness model inputs

Variable	Mean	Low CI/Q/n	Upper CI/Q/N	PSA	Source
Patient characteristics					
Primary care age	25	..	..	Deterministic	CG61 assumption
Primary care female	50.0%	..	..	Deterministic	CG61 assumption
Secondary care age	16	..	..	Deterministic	Assumption
Secondary care female IBD	38.5%	35	91	Beta	Henderson et al 2012
Secondary care female non-IBD	44.4%	44	99	Beta	Henderson et al 2012
Disease prevalences and SMRs					
Primary care IBD prevalence	6.3%	7	111	Beta	Durham data
Primary care CD:UC	40.4%	365	904	Beta	Shivananda et al 1996
Secondary care IBD prevalence	47.9%	91	190	Beta	Henderson et al 2012
Secondary care CD:UC split	74.7%	62	83	Beta	Henderson et al 2012
CD SMR	1.38	1.23	1.55	Lognormal	Bewtra, M. et al 2013
UC SMR	1.19	1.00	1.35	Lognormal	Bewtra, M. et al 2013
IBS SMR	1.00	..	..	..	Assumption
Test accuracies					
GP sensitivity	100%	7	7	Beta	Otten et al 2008
GP specificity	78.8%	82	104	Beta	Otten et al 2008
CalDetect 15µg/g sensitivity	100.0%	23	23	Beta	Otten et al 2008
CalDetect 15µg/g specificity	94.5%	86	91	Beta	Otten et al 2008
CalDetect 60µg/g sensitivity	60.9%	14	23	Beta	Otten et al 2008
CalDetect 60µg/g specificity	97.8%	89	91	Beta	Otten et al 2008
CalDetect 15µg/g sensitivity	95.7%	22	23	Beta	Hessells et al 2012
CalDetect 15µg/g specificity	53.2%	33	62	Beta	Hessells et al 2012
CalDetect 60µg/g sensitivity	87.0%	20	23	Beta	Hessells et al 2012
CalDetect 60µg/g specificity	74.2%	46	62	Beta	Hessells et al 2012
Quantum Blue 30µg/g sensitivity	95.7%	22	23	Beta	Hessells et al 2012
Quantum Blue 30µg/g specificity	69.4%	43	62	Beta	Hessells et al 2012
Quantum Blue 40µg/g sensitivity	91.3%	21	23	Beta	Hessells et al 2012
Quantum Blue 40µg/g specificity	83.9%	52	62	Beta	Hessells et al 2012
Quantum Blue 50µg/g sensitivity	87.0%	20	23	Beta	Hessells et al 2012
Quantum Blue 50µg/g specificity	83.9%	52	62	Beta	Hessells et al 2012
Quantum Blue 60µg/g sensitivity	78.3%	18	23	Beta	Hessells et al 2012
Quantum Blue 60µg/g specificity	87.1%	54	62	Beta	Hessells et al 2012

Elisa 50µg/g sensitivity IBD vs IBS	93.0%	83.0%	97.0%	Gamma detriment	EAG NMA fig. 6
Elisa 50µg/g specificity IBD vs IBS	94.0%	73.0%	99.0%	Gamma detriment	EAG NMA fig. 6
Elisa 50µg/g sensitivity IBD vs non-IBD	93.0%	83.0%	97.0%	Gamma detriment	EAG NMA fig. 13
Elisa 50µg/g specificity IBD vs non-IBD	94.0%	73.0%	99.0%	Gamma detriment	EAG NMA fig. 13
Elisa 100µg/g sensitivity IBD vs non-IBD	93.0%	83.0%	97.0%	Gamma detriment	EAG NMA fig. 16
Elisa 100µg/g specificity IBD vs non-IBD	94.0%	73.0%	99.0%	Gamma detriment	EAG NMA fig. 16
Elisa 50µg/g sensitivity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 50µg/g specificity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 100µg/g sensitivity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 100µg/g specificity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 150µg/g sensitivity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 150µg/g specificity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 50µg/g sensitivity IBD vs non-IBD	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 50µg/g specificity IBD vs non-IBD	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 100µg/g sensitivity IBD vs non-IBD	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Elisa 100µg/g specificity IBD vs IBS	[redacted]	[redacted]	[redacted]	Beta	Basumani et al 2013
Colonoscopy sensitivity	95.0%	..	..	Deterministic	Expert opinion
Colonoscopy specificity	100.0%	..	..	Deterministic	Expert opinion
IBS and IBD false negatives					
IBS response to dietary advice	47.0%	33.0%	57.0%	LogNormal	Mearin 2004
IBS subsequence non-responders to medication	5.0%	..	..	Deterministic	YHEC expert opinion
IBD FN response to dietary advice	0%	..	..	Deterministic	Assumption
IBD FN subsequence non-responders to medication	100.0%	..	..	Deterministic	Assumption
Time to representation among non-responders (weeks)	12	..	..	Deterministic	Expert opinion
Treatment effectiveness CD					
CD induction therapy					
Prednisolone duration weeks	8	..	..	Deterministic	CG61 assumption
Prednisolone response rate	67.13%	..	..	Lookup table	CG61 NMA
Prednisolone withdrawal rate	11.83%	..	..	Lookup table	CG61 NMA
Prednisolone + azathioprine duration weeks	8	..	..	Deterministic	CG61 assumption
Prednisolone + azathioprine response rate	65.74%	..	..	Lookup table	CG61 NMA
Prednisolone + azathioprine withdrawal rate	9.77%	..	..	Lookup table	CG61 NMA
Adalimumab duration weeks	6	..	..	Deterministic	CG61 assumption
Adalimumab response rate	62.34%	..	..	Lookup table	CG61 NMA
Adalimumab withdrawal rate	10.47%	..	..	Lookup table	CG61 NMA
CD induction therapy responders to no treatment	20.0%	..	..	Deterministic	Expert opinion

CD maintenance therapy					
Azathioprine duration (repeated/cycle length) weeks	8			Deterministic	CG61 assumption
Azathioprine withdrawal to no treatment (conservative)	0.00%			Deterministic	CG61 assumption
Azathioprine relapse to induction (conservative)	5.33%	..	..	Lookup table	CG61 NMA
CD post maintenance induction therapy					
Prednisolone duration weeks	8	..	..	Deterministic	CG61 assumption
Prednisolone response rate	59.19%	..	..	Lookup table	CG61 NMA
Prednisolone + azathioprine duration weeks	8	..	..	Deterministic	CG61 assumption
Prednisolone + azathioprine response rate	59.31%	..	..	Lookup table	CG61 NMA
Adalimumab duration weeks	6	..	..	Deterministic	CG61 assumption
Adalimumab response rate	55.81%	..	..	Lookup table	CG61 NMA
Adalimumab maintenance of response early thereafter (weekly)	87.12%	..	..	Lookup table	CG61 NMA
Adalimumab maintenance of response subsequent (weekly)	96.37%	..	..	Lookup table	CG61 NMA
Treatment effectiveness UC					
UC induction therapy					
Low dose ASA duration weeks	8	..	..	Deterministic	UC guideline assumption
Low dose ASA withdrawal	10.9%	..	..	Lookup table	UC guideline NMA
Low dose ASA response rate | no withdrawal	33.4%	..	..	Lookup table	UC guideline NMA
High dose ASA duration weeks	8	..	..	Deterministic	UC guideline assumption
High dose ASA withdrawal	9.1%	..	..	Lookup table	UC guideline NMA
High dose ASA response rate | no withdrawal	44.1%	..	..	Lookup table	UC guideline NMA
High dose ASA + topical duration weeks	4	..	..	Deterministic	UC guideline assumption
High dose ASA + topical withdrawal	11.1%	..	..	Lookup table	UC guideline NMA
High dose ASA + topical response rate | no withdrawal	52.1%	..	..	Lookup table	UC guideline NMA
High dose ASA + beclometasone duration weeks	4	..	..	Deterministic	UC guideline assumption
High dose ASA + beclometasone withdrawal	2.3%	..	..	Lookup table	UC guideline NMA
High dose ASA + beclometasone response rate | no withdrawal	67.6%	..	..	Lookup table	UC guideline NMA
Prednisolone duration weeks	8	..	..	Deterministic	UC guideline assumption
Prednisolone withdrawal	0.0%	..	..	Lookup table	UC guideline assumption
Prednisolone response | no withdrawal	52.4%	..	..	Lookup table	UC guideline NMA
Inpatient duration weeks	1	..	..	Deterministic	UC guideline assumption
Inpatient withdrawal	0.0%	..	..	Deterministic	UC guideline assumption
Inpatient response | no withdrawal	91.0%	..	..	Deterministic	UC guideline assumption
Surgery duration weeks	1	..	..	Deterministic	UC guideline assumption
Surgery duration response	100.00%	..	..	Deterministic	UC guideline assumption
UC induction therapy responders to no treatment	20.0%	..	..	Deterministic	Expert opinion

UC maintenance therapy					
Maintenance duration (repeated / cycle lengh) weeks	8	..	..	Deterministic	UC guideline assumption
LASA withdrawal	9.7%	..	..	Lookup table	UC guideline NMA
LASA loss or remission | no withdrawal	6.8%	..	..	Lookup table	UC guideline NMA
Azathioprine loss of remission	5.9%	..	..	Lookup table	UC guideline NMA
No therapy loss of remission	13.5%	..	..	Lookup table	UC guideline NMA
UC Induction post maintenance therapy					
As for induction therapy	..	..	..	Lookup table	UC guideline NMA
Administration costs per course					
Prednisolone – derived from multiple NHS reference costs	£164	..	..	LogNormals	NHS reference costs 2011-12
Azathioprine – derived from multiple NHS reference costs	£164	..	..	LogNormals	NHS reference costs 2011-12
Adalimumab – derived from multiple NHS reference costs 6 wk	£241	..	..	LogNormals	NHS reference costs 2011-12
Adalimumab – derived from multiple NHS reference costs 8 wk	£280	..	..	LogNormals	NHS reference costs 2011-12
LASA Induction	£95			LogNormals	NHS reference costs 2011-12
HASA Induction	£95			LogNormals	NHS reference costs 2011-12
HAST Induction	£50			LogNormals	NHS reference costs 2011-12
HASB Induction	£50			LogNormals	NHS reference costs 2011-12
LASA Maintenance	£24			LogNormals	NHS reference costs 2011-12
Azathioprine Maintenance	£24			LogNormals	NHS reference costs 2011-12
Drug costs per course					
Prednisolone – derived from multiple NHS reference costs	£38.10			Deterministic	BNF March 2013
Azathioprine – derived from multiple NHS reference costs	£42.92			Deterministic	BNF March 2013
Adalimumab – derived from multiple NHS reference costs 6 wk	£1408.56			Deterministic	BNF March 2013
Adalimumab – derived from multiple NHS reference costs 8 wk	£1760.70			Deterministic	BNF March 2013
LASA Induction	£74.24			Deterministic	BNF March 2013
HASA Induction	£152.07			Deterministic	BNF March 2013
HAST Induction (4 week)	£180.57			Deterministic	BNF March 2013
HASB Induction (4 weeks)	£132.59			Deterministic	BNF March 2013
LASA Maintenance	£74.24			Deterministic	BNF March 2013
Azathioprine Maintenance	£42.92			Deterministic	BNF March 2013
Utilities					
CD remission	0.890	s.d. 0.130 n=129		LogNormal	Stark et al
CD no remission	0.610	s.d. 0.290 n=97		LogNormal	Stark et al
UC remission	0.910	s.d. 0.140 n=138		LogNormal	Stark et al
UC no remission	0.710	s.d. 0.180 n=81		LogNormal	Stark et al
IBS remission – no remission increment	0.071	0.020	0.147	LogNormal	CG 61

IBS no remission	0.662	..	..	Deterministic	Brazier et al 2004
Test staff timings and costs					
Staff time GP nurse CalDetect minutes	15.00	..	..	Deterministic	Expert opinion
Staff time Grade 6/7 Quantum Blue minutes	12.50	..	..	Deterministic	Expert opinion
Staff time Grade 6/7 ELISA minutes	11.75	..	..	Deterministic	Expert opinion
Staff time GP nurse CalDetect cost	£8.32	..	..	Deterministic	Expert opinion
Staff time Grade 6/7 Quantum Blue cost	£8.65	..	..	Deterministic	Expert opinion
Staff time Grade 6/7 ELISA cost	£8.13	..	..	Deterministic	Expert opinion
GP per appointment	£36.00			Deterministic	PSSRU Unit costs
Other costs					
301 OP Consultant First Face to Face non-admitted	£164	£113	£194	LogNormal	NHS reference costs 2011-12
301 OP Consultant Follow-Up Face to Face non-admitted	£115	£79	£142	LogNormal	NHS reference costs 2011-12
301 OP Nurse Follow-Up Face to Face non-admitted	£85	£65	£101	LogNormal	NHS reference costs 2011-12
DAP823 Haematology	£3.09	£1.76	£4.18	LogNormal	NHS reference costs 2011-12
DAP831 Virology	£7.75	£5.25	£9.99	LogNormal	NHS reference costs 2011-12
DAP841 Biochemistry	£1.23	£0.80	£1.46	LogNormal	NHS reference costs 2011-12
FZ51Z OP Colonoscopy no biopsy	£276.32	£219.30	£306.16	LogNormal	NHS reference costs 2011-12
FZ52Z OP Colonoscopy with biopsy	£316.92	£283.18	£309.29	LogNormal	NHS reference costs 2011-12
FZ51Z Day case Colonoscopy no biopsy	£527.24	£413.47	£577.85	LogNormal	NHS reference costs 2011-12
FZ52Z Day case Colonoscopy with biopsy	£570.45	£449.52	£657.86	LogNormal	NHS reference costs 2011-12
FZ54Z OP Sigmoidoscopy no biopsy	£174.05	£110.68	£224.91	LogNormal	NHS reference costs 2011-12
FZ55Z OP Sigmoidoscopy with biopsy	£169.84	£91.79	£214.97	LogNormal	NHS reference costs 2011-12
FZ54Z Day case Sigmoidoscopy no biopsy	£445.88	£335.10	£508.42	LogNormal	NHS reference costs 2011-12
FZ55Z Day case Sigmoidoscopy with biopsy	£480.96	£373.35	£544.25	LogNormal	NHS reference costs 2011-12
FZ37G Inpatient non-surgical ellective	£4,092.94	£1,648.26	£5,197.01	LogNormal	NHS reference costs 2011-12
FZ37H Inpatient non-surgical ellective	£2,570.84	£1,546.94	£3,173.01	LogNormal	NHS reference costs 2011-12
FZ37I Inpatient non-surgical ellective	£2,574.02	£1,405.67	£3,106.13	LogNormal	NHS reference costs 2011-12
FZ37J Inpatient non-surgical ellective	£1,981.62	£1,327.26	£2,341.05	LogNormal	NHS reference costs 2011-12
FZ74A Inpatient surgical	£8,281.13	£6,646.52	£9,666.87	LogNormal	NHS reference costs 2011-12
FZ74A Inpatient surgical	£6,127.84	£5,148.80	£6,855.26	LogNormal	NHS reference costs 2011-12