Gastroenterol Hepatol. 2019;42(4):263–270

Gastroenterología y Hepatología

www.elsevier.es/gastroenterologia

REVIEW

Faecal immunochemical tests for haemoglobin (FIT) in the assessment of patients with lower abdominal symptoms: current controversies

Callum G. Fraser

Centre for Research into Cancer Prevention and Screening, University of Dundee, Ninewells Hospital and Medical School, Dundee, Scotland, UK

Received 14 August 2018; accepted 21 September 2018

KEYWORDS
Colorectal cancer;
Faecal haemoglobin;
Faecal immunochemical test;
Faecal occult blood test;
Lower abdominal symptoms;
Significant bowel disease

Abstract Faecal immunochemical tests for haemoglobin (FIT), as an adjunct to clinical information, assist in the triage of patients presenting in primary care with lower abdominal symptoms. Controversy remains regarding whether and which qualitative and quantitative FIT can be used, which groups of patients would benefit most from FIT, whether FIT should be done in primary and/or secondary care, and how FIT should be incorporated into diagnostic pathways. Controversy also exists as to the optimum cut-off used for referral for colonoscopy. A single sample of faeces may be sufficient. Reporting of results requires consideration. FIT provide a good rule in test for colorectal cancer and a good rule out test for significant bowel disease, but robust safety-netting is required for patients with negative results and ongoing symptoms. Risk scoring models have been developed, but their value is unclear as yet. Further evaluation of these topics is required to inform good practice.

PALABRAS CLAVE
Cáncer colorrectal;
Hemoglobina fecal;
Prueba inmunoquímica

Pruebas inmunoquímicas fecales (PIF) para determinar la hemoglobina en la evaluación de pacientes con síntomas abdominales inferiores: controversias actuales

Resumen Las pruebas inmunoquímicas fecales (PIF) para determinar la hemoglobina, como pruebas complementarias de la información clínica, ayudan a la selección de pacientes que acuden a Atención Primaria con síntomas abdominales inferiores. Sin embargo, continúa la

Abbreviations: AA, advanced adenoma; AN, advanced neoplasia; ACRN, advanced colorectal neoplasia; AUC, area under the receiver operating characteristic (ROC) curve; CRC, colorectal cancer; f-Hb, faecal haemoglobin concentration; FIT, faecal immunochemical test for haemoglobin; Hb, haemoglobin; HGD, high grade dysplasia; IBD, inflammatory bowel disease; LoD, limit of detection; LoQ, limit of quantitation; POC, point of care; PPV, positive predictive value; SBD, significant bowel disease.

E-mail addresses: callum.fraser@nhs.net, c.g.fraser@dundee.ac.uk

fecal;
Prueba de sangre oculta en materia fecal;
Síntomas abdominales inferiores;
Enfermedad intestinal importante

polémica sobre si utilizar PIF y, en ese caso, si utilizar PIF cualitativas o cuantitativas, qué grupos de pacientes se beneficiarían más de las PIF, si las PIF deben realizarse en Atención Primaria o en el especialista, y cómo las PIF deben incorporarse a los protocolos de diagnóstico. También hay polémica sobre el valor de corte óptimo utilizado para la derivación a colonoscopia. Una sola muestra de heces puede ser suficiente. La notificación de los resultados exige análisis. Las PIF son un buen principio en la prueba del cáncer colorrectal y una buena prueba para descartar una enfermedad intestinal importante, pero se necesita una red de protección sólida para los pacientes con resultados negativos y síntomas continuos. Se han elaborado modelos de puntuación de riesgo, pero su valor aún no está claro. Se requiere una evaluación adicional de estos temas para lograr una buena práctica.

Introduction

Faecal immunochemical tests for haemoglobin (FIT) are now widely used in asymptomatic colorectal (bowel) cancer screening since widely perceived as the best currently available non-invasive approach: participants with positive results are referred for further investigation, usually colonoscopy.[1] Possibly because of (a) the positive publicity surrounding such programmes, (b) the information given that participants who experience lower abdominal symptoms between screening episodes should seek medical care and (c) the influence of well-known individuals in the media,[2] the demand for colonoscopy has risen further over recent times. In addition, because more bowel disease is found, an increased requirement for colonoscopic surveillance after treatment has occurred, although there are early indications that FIT might be useful in this clinical setting as well as others.[3] Further, there have been local, regional and national efforts in many countries encouraging people with abdominal symptoms to seek urgent medical attention in primary care, again leading to further demands on colonoscopy services.[4]

However, colonoscopy is often a limited resource and, in consequence, it would be of much benefit if a simple, inexpensive investigation would aid in deciding which of the many patients presenting with symptoms, particularly in primary care, would benefit from colonoscopy and which would not. This is particularly germane since, as pointed out some time ago by Jellema et al.[5] and very well documented more recently by Vega et al.,[6] diagnosis is a challenge, since there are often no specific symptoms and lower abdominal symptoms are very common and mostly related to problems other than significant bowel disease (SBD), which includes colorectal cancer (CRC), advanced adenoma (AA) that are sometimes precursors of CRC, and inflammatory bowel disease (IBD). Indeed, according to the National Institute for Health and Care Excellence (NICE) in England, common symptoms have a positive predictive value (PPV) for CRC of only 3–4%.[7] Thankfully, there is ever growing evidence that FIT can be used to assist in the triage of patients presenting with symptoms of lower bowel disease.

FIT in assessment of patients with lower abdominal symptoms

The evidence for the successful application of FIT in assessment of patients presenting in primary care with lower abdominal symptoms, and patients being seen in secondary care clinics after referral, has been very well documented in a number of recent reviews.[8–11] Therefore, the fine details of the individual studies will not be repeated in detail in this review. Although it is known that there are a number of real-world pilots, feasibility studies and evaluations of FIT in the assessment of symptomatic patients underway and publications on this topic are in press or in preparation, there appears to have been only one further relevant publication[12] since the most recent comprehensive review.[11] This study investigated the value of a quantitative FIT in the diagnostic process of CRC and other SBD in individuals presenting with low risk symptoms in general practice, FIT being used as a rule-in test on patients aged $\geq$30 years with the faecal haemoglobin concentration (f-Hb) cut-off for referral to colonoscopy being 10 μg Hb/g faeces: 3462 FIT were performed and 540 (15.6%) had positive results. Of these, 51 (PPV: 9.4%) individuals with a positive FIT result were diagnosed with CRC and 73 (PPV: 13.5%) with other SBD. The false negative test result rate for CRC was < 0.1%. It was concluded that FIT may be used as a supplementary diagnostic test in general practice in the diagnostic process of CRC and other SBD in individuals with low risk of CRC.

Even though it has been documented by NICE,[13] a highly respected developer of guidelines, that quantitative FIT are recommended for adoption in primary care to guide referral for suspected CRC in people without rectal bleeding who have unexplained symptoms but do not meet the criteria for a suspected cancer pathway referral outlined in the NICE NG12 guideline,[7] and that results should be reported using a f-Hb cut-off of 10 μg Hb/g faeces, many controversies still exist: a number of these will be addressed in this review.

Qualitative or quantitative FIT

FIT are available in two formats.[14] Qualitative FIT give a dichotomous, positive/negative result, usually using

Table 1 Advantages and disadvantages of qualitative and quantitative faecal immunochemical tests for haemoglobin (FIT).

Qualitative FIT
Advantages
(a) supposedly simple to perform;
(b) inexpensive, even in small numbers;
(c) no need for analytical instrumentation;
(d) can be done by others in health care than professionals in laboratory medicine;
(e) easy to store since no refrigeration of components needed;
(f) no calibration is needed;
(g) integral quality monitors are always present; and
(h) results are available for clinical decision-making within minutes.
Disadvantages
(a) it is not simple to interpret faint lines on strips or cassettes;
(b) colour develops dynamically and true negatives become positive with a prolonged read time and results may be falsely negative if read too early;
(c) no quality control with an appropriate matrix is possible;
(d) difficult and time consuming to do large numbers;
(e) no automation generally possible, although some small readers are available;
(f) usually impossible to download data directly resulting in possibilities of transcription mistakes;
(g) f-Hb cut-off concentrations are set by the manufacturer, are not the same for different FIT and it is difficult to decide which to use in practice; and
(h) lot-to-lot variation is possible and some acceptance quality checks are needed to confirm cut-off.
Quantitative FIT
Advantages
(a) high quality analyses with good reproducibility;
(b) easy to monitor quality using total quality management techniques to guarantee standards through International Standardisation Organisation (ISO) 15198 accreditation;
(c) high throughput of samples may be possible;
(d) no visual interpretation of results;
(e) download of data into LIS via middleware may be possible, eliminating transcription errors and facilitating record keeping; and
(f) linkage with other data, for example, age and sex, may be possible and may be important for the future for risk scoring or monitoring; and
(g) the cut-off f-Hb for referral for colonoscopy can be set locally.
Disadvantages
(a) expensive if few FIT analyses done;
(b) need for instrumentation, installation, training, etc.;
(c) the need to evaluate or validate for laboratory medicine accreditation systems and then prepare complex documentation;
(d) difficult to decide which FIT analytical system to use since many now available;
(e) only done by trained professionals in laboratory medicine;
(f) refrigeration required for latex reagent and quality control materials and calibrators; and
(g) issues around stability of haemoglobin in specimen collection devices.

lateral-flow immunochromatographic cassettes or strips. Quantitative FIT usually involve automated immunoturbidimetry on small bench-top dedicated analytical systems and provide a numerical estimate of the f-Hb: such examinations can also be performed on larger routine laboratory medicine automated systems. As detailed previously,[10,11] both constructs have been used to evaluate the use of FIT in assessment of patients with symptoms. Some of the advantages and disadvantages of qualitative and quantitative FIT are listed in Table 1.

Selecting a faecal immunochemical test

Although there are a large number of qualitative[15] and qualitative[16] FIT available, it could be argued that the available evidence suggests that both qualitative and quantitative FIT appear to give somewhat similar clinical outcomes[10] and both could be used in assessment of patients presenting with lower abdominal symptoms if the disadvantages of the two rather different approaches were carefully kept in mind. However, it must be noted

that the data available to support the use of FIT in this clinical setting was generated with only very few of the qualitative[15] and quantitative[16] FIT available. There are more data on quantitative than qualitative FIT and only the former were recommended by NICE.[13] In consequence, it has been advocated that assessment of the benefits and harms of qualitative FIT being done by people in their own homes should be investigated,[17] as should the use in general practice and in secondary healthcare settings, particularly in gastroenterology and other clinics evaluating patients before colonoscopy.[10,11]

It is important to note that the numerical results generated using different FIT may not be the same. Three quantitative FIT systems were recommended in NICE DG30, namely the OC-Sensor (Eiken Chemical Co. Ltd., Tokyo, Japan), the HM-JACKarc (Kyowa Medex Co., Ltd., Tokyo, Japan) and FOB-Gold (Sentinel Diagnostics, Milan, Italy)[13]; however, the evidence gathered to support the recommendation of these three FIT systems clearly does show differences in the clinical outcomes obtained. The main reason for the differences is probably that polyclonal antibodies are used, which may react differently to the spectra of intact haemoglobin (Hb) and early degradation products present in faecal samples collected for analysis. In addition, the analytical performance characteristics of the FIT systems show a number of marked differences, for example, the analytical range, the limit of detection, the lowest f-Hb that can be reliably distinguished from a sample with no f-Hb, and the limit of quantitation, the lowest f-Hb that can be reliably measured, differ between these three quantitative FIT systems.[18] In addition, there are a number of pre-analytical and analytical aspects which differ between FIT and these include lack of harmonisation of the specimen collection devices, the intrinsic heterogeneity and variable consistency of faecal samples and the effect of these variables on faecal collection, the different recommended techniques for collection of faeces and the fact that no primary reference material or method is currently available to standardise FIT.[19] Further, a very relevant concern is the transferability of already published data over time and geography: manufacturers of FIT continually evolve their products and, in consequence, outcomes may not be comparable over time. An example of change over time is that two of the quantitative FIT recommend by NICE[13] have had improvements made to their buffers, supposedly to increase the stability of any haemoglobin present prior to analysis.[20,21]

Although there are a number of studies comparing different FIT in asymptomatic population-based screening applying different strategies,[22] which do demonstrate differences between systems when the same cut-off f-Hb is used for referral to colonoscopy, which are minimised if the same positivity is used,[23–25] there seem no head to head comparisons of FIT in assessment of patients with symptoms. This would be of considerable interest, since the same FIT are used in screening and assessment of the symptomatic and it has been amply demonstrated[15,16] that there are many differences between available FIT used in screening.

Deciding which patients should undertake a FIT

The clinical outcome characteristics will depend very much on the type of patients who have provided faecal samples for FIT analysis. The spectra of patients examined have varied from study to study.[10,11] NICE DG30 specifically states,[13] as detailed above, that FIT are to be applied in what might be termed low risk patients and those patients who have rectal bleeding and meet the criteria for a suspected cancer pathway referral outlined in NICE NG12[7] should be referred for further investigation without FIT. However, most evaluations have assessed the value of FIT in quite mixed populations including those at high risk of CRC and even those undergoing surveillance for previous disease.[26] In addition, most studies, except for that of Mowat et al.[27] and Juul et al.[12] have not evaluated the use of FIT in primary care, but have examined patients who were already referred for colonoscopy through a variety of clinical pathways with different criteria for referral. Further studies on the routine application of FIT in primary care are urgently required, since this seems to be the most appropriate sector in which FIT should be requested. However, studies on the appropriate uses of FIT in secondary care, such as in gastroenterology clinics, would also be of considerable interest.

In addition, there has been some controversy about how FIT should be integrated into diagnostic pathways. One study, following on directly from an evaluation of point of care (POC) FIT and calprotectin in patients with symptoms,[28] developed a multivariable diagnostic model for SBD with routine clinical information and subsequently extended this with faecal calprotectin testing and/or qualitative POC FIT.[29] The results were said to underscore that a positive f-Hb result already implies the need for referral and that clinical data do not add much. However, it was suggested that these data are informative when the f-Hb result is negative. It was concluded that a diagnostic strategy with routine clinical data and f-Hb alone may safely rule out SBD and prevent unnecessary endoscopy referral in approximately one-third of patients. A contrasting editorial suggested that a single quantitative f-Hb result, without any clinical information, could be sufficient to decide whom to refer for colonoscopy and, because of the significant overlap of symptoms in those with and without SBD, could be the primary investigation performed.[30] This thesis has been supported in a recent study which compared the utility of FIT as the initial investigation with the original 2015 NICE NG12 symptom based guidelines.[31] Data from three studies done in Scotland were included and overall diagnostic accuracy was also estimated by the area under the receiver operating characteristic (ROC) curve (AUC). The AUC for CRC was 0.85 for FIT versus 0.65 for NG12 and, for SBD, the AUC was 0.73 for FIT versus 0.56 for NG12. It was concluded that f-Hb provided a good rule-out test for SBD and had significantly higher overall diagnostic accuracy than the 2015 NG12 guidelines.[7]

Other approaches are possible[10] and, according to a recent review on setting up a service for FIT for assessment of symptomatic patients, there appears to be no ''best

practice'' that can be detailed at this particular time[19]: reports on the results of the use of FIT for the routine assessment of patients in primary care are awaited with interest, but informal current consensus seems to be that the f-Hb found should be taken into account along with symptoms and clinical findings, particularly chronic diarrhoea and the presence of an abdominal mass, and the results of the full blood count, particularly the detection of anaemia, as recently advocated by Hogberg et al.[32]

Selecting the cut-off faecal haemoglobin concentration (f-Hb) used for referral for colonoscopy

A number of the evaluations of the use of quantitative FIT in assessment of the symptomatic have explored the relationship between f-Hb and clinical outcomes. Since it is very well documented that f-Hb is directly related to the severity of colorectal disease, and as confirmed recently,[33] it is not surprising that, as the f-Hb cut-off is lowered, the sensitivity increases and the specificity decreases. A good example is provided in a supplementary table in the study of Rodríguez-Alonso et al.,[34] where, at f-Hb cut-offs > 0, ≥10, ≥15 and ≥20 μg Hb/g faeces, sensitivity for CRC was 100, 96.7, 96.7 and 93.3% with specificity of 43.3, 79.8, 83.1 and 86.1% and, for advanced neoplasia (AN: CRC + AA), sensitivity was 81.2, 61.7, 57.1 and 53.4%, while specificity was 45.5, 83.4, 86.6 and 89.4%. It is important to note that, at 20 μg Hb/g faeces, a commonly used cut-off in asymptomatic population-based screening, and as used by Cubiella et al.,[35] a small number of cases of CRC are missed. This has also been shown for AA and IBD at f-Hb cut-offs of 10 μg Hb/g faeces, as recommended by McDonald et al.[36] and Godber et al.[37] This missing of cases of SBD was also found by Mowat et al.[27] with a f-Hb cut-off of 10 μg Hb/g faeces, but who also investigated ''detectable'' f-Hb as a cut-off (defined as >0 μg Hb/g faeces) achieving sensitivities of 100, 82.5 and 85.3% for CRC, AA and IBD, respectively. Similarly, Widlak et al. stated that an undetectable FIT haemoglobin is sufficiently sensitive to exclude CRC, but they defined ''undetectable'' as f-Hb <7 μg Hb/g faeces, the LoQ of the FIT system used.[38]

The publications on FIT in the assessment of patients with symptoms clearly demonstrate the current controversy of which f-Hb cut-off to use and also the dilemma of how to report the results of FIT analyses. This has been addressed in detail recently[39] and a series of proposals made including the following[40]:

- f-Hb concentrations should not be reported to more significant figures than whole integers,
- f-Hb concentrations less than the limit of detection (LoD) of the FIT analytical system should be termed ''not detected'' or ''undetectable'',
- for academic use: f-Hb greater than the LoD could advantageously be documented,
- for routine clinical use: numerical f-Hb should be reported only when greater than the limit of quantitation (LoQ): f-Hb less than the LoQ (x) should be reported as $<x$ μg Hb/g faeces and
- if a more sophisticated reporting system is required, one option is to report:
  - f-Hb < LoD = f-Hb not detected
  - LoD < f-Hb < LoQ = f-Hb detected
  - f-Hb ≥ LoQ = report the found f-Hb

Adherence to these proposals would result in harmonisation of the reporting of f-Hb data, which should facilitate understanding and transferability of information across geography and time.

A further controversy is whether the high sensitivity makes FIT a good rule-in test for CRC or the high negative predictive value (NPV) shown in many studies on use of FIT in assessment of the symptomatic[10,11] demonstrates that FIT provides a good rule out investigation for SBD. In reality, f-Hb in patients presenting in primary carer with lower abdominal symptoms could be considered as a continuous variable. The most important use in colonoscopy constrained countries is likely to be to stop patients with vague symptoms and unlikely to have SBD being referred for colonoscopy. If the f-Hb is lower than the selected f-Hb cut-off and if the patient does not have what are sometimes termed red flag symptoms, then the risk of SBD is small: however, this does not simply mean informing the patient nothing is wrong. Many lower abdominal symptoms are transient, so it might be satisfactory to leave these patients without any immediate further investigation but, for others who continue to have symptoms, it is obligatory to have robust safety-netting procedures in place as recently described in a review of FIT in patients with symptoms,[41] including watching and waiting, referral to gastroenterology in secondary care, and perhaps a repeat FIT, although there is no evidence to date that this is useful. This is important since, as discussed above, a few cases of CRC may be missed in addition to a few more cases of AA and IBD. Moreover, since it is well documented that f-Hb is related to disease severity,[33] the higher the f-Hb, the greater the risk of SBD. In consequence, those with very high f-Hb might benefit from more rapid referral to colonoscopy than those who have slightly elevated f-Hb.

How many faecal samples should be taken for FIT?

It is dogma that some neoplastic lesions in the colon bleed intermittently and so it is also dogma that more than one faecal sample should be collected. Although there is considerable literature on one versus two samples in asymptomatic population screening, the literature on the effect of number of FIT samples in assessment of patients with symptoms is sparse. A recent study assessed the use of three samples in patients with CRC and adenomas with high grade dysplasia (HGD) that initially presented with symptoms to primary care and completed FIT.[42] Of 195 patients, 160 delivered three FIT. Using the 139 cases in which at least one sample was positive, the likelihood of detecting a positive sample upon analysis of only one of the three samples was 0.91, indicating that 13 positive cases may have been missed. It was concluded that use of one sample instead of three samples may result in missing about 10% of symptomatic CRC and adenomas with HGD. Unfortunately, this study was done with a visually read qualitative FIT and dipstick test, with f-Hb cut-off of 25–50 μg Hb/g faeces, rather higher than f-Hb cut-offs used in the published studies using

quantitative FIT. It may be unsurprising, therefore, that cases of SBD were missed. Auge et al.[26] examined the clinical utility of one versus two samples for FIT samples in the detection of advanced colorectal neoplasia (ACRN: CRC + AA) in symptomatic patients using a quantitative FIT. It was found that the diagnostic yield, when two samples for FIT were collected (using f-Hb cut-off of 20 μg Hb/g faeces), could be achieved with one sample, albeit using a lower f-Hb cut-off (10 μg Hb/g faeces). With a different FIT analytical system, using two samples for each patient and choosing the highest result, the sensitivity for ACRN was 40.0%, with a specificity of 88.6%, and a similar diagnostic yield was again obtained using only one sample and decreasing the f-Hb cut-off.[43] Moreover, when one sample and a 10 μg Hb/g faeces cut-off was used, it was possible to rule out the majority of malignant lesions. Based on this small amount of evidence, since collecting multiple samples involves more funding and effort and might decrease the acceptability of the test for patients, it is likely that one sample combined with a low f-Hb cut-off would provide a cost-effective and clinically efficient service for patients presenting in primary care.

FIT alone or in combination with other variables

Risk prediction models which take both symptoms and multiple risk factors into account might have potential to improve timely diagnosis of SBD. Williams et al.[44] have systematically identified and compared the performance of models that predict the risk of primary CRC among symptomatic individuals: it was concluded that good approaches had been generated in both primary and secondary care populations. Most were said to contain variables that were easily obtainable in a single consultation. However, few of the models actually include f-Hb in the algorithm. Since it is well documented that f-Hb increase with age and is higher in men than women,[45] although the actual f-Hb vary from country to country,[46] Rodríguez-Alonso et al.[34] created a simple risk score for AN based upon age, gender and f-Hb. The points attributed to each risk factor were weighted according to their respective coefficients in a multiple logistic regression model and the score had a range of 0–11 points based on the sum of the points in the individual patient. A simple chart to calculate the sum was published. In the population studied, if a risk score $\geq$ 5 was considered as the referral criterion for colonoscopy, only 36.4% of would be referred; no cases of CRC and only 5% of AA were undetected. A more complex approach was developed by Cubiella et al.[47] using a multivariate logistic regression analysis to develop the model, with diagnostic accuracy of CRC detection as the main outcome: 1572 symptomatic patients were included in the derivation cohort and 1481 in the validation cohorts. The final prediction model included 11 variables: age, male sex, f-Hb $\geq$20 μg Hb/g faeces, blood Hb <10 g/dl, blood haemoglobin 10–12 g/dl, carcinoembryonic antigen $\geq$3 ng/ml, aspirin use, previous colonoscopy, presence of a rectal mass, benign anorectal lesion, rectal bleeding, and change in bowel habit. The AUC was 0.92. On the basis of the thresholds with 90% and 99% clinical sensitivity, the derivation cohort was divided into high, intermediate and low risk groups for CRC with PPV of 40.7, 4.4 and 0.2% respectively. It was concluded that the COLONPREDICT strategy developed was a highly accurate prediction model for CRC detection. Because of the complexity of this model, Cubiella et al. went on to develop the faecal haemoglobin concentration, age and sex test (FAST) score with data from five diagnostic test accuracy studies that evaluated quantitative FIT in symptomatic patients referred for colonoscopy[48]: 1572 and 3976 patients were examined in derivation and validation cohorts, respectively. The AUC for CRC detection was 0.88 and 0.91 in the derivation and validation cohorts. The FAST score was said to an easy to calculate prediction tool, highly accurate for CRC detection in symptomatic patients. Other variables affect f-Hb, such as deprivation,[49,50] and it may be that these should be examined for incorporation into future risk scoring strategies. However, it is clear that further research is needed to assess the clinical utility of these risk scoring strategies and other complex approaches before they can be incorporated into routine practice.

Conclusions

Although there is much evidence from research studies that FIT, as an adjunct to clinical information and full blood count, can provide a very useful tool to assist with the triage of patients presenting in primary care with lower abdominal symptoms, there remain a number of issues which can only be resolved through further study and evaluation as FIT become more and more applied in real-world clinical practice.[51] There remains some controversy regarding whether and which qualitative and quantitative FIT can be applied in this clinical setting, which particular groups of patients would benefit from FIT, and whether the investigation should be done in primary and/or secondary care settings and how it should be incorporated into diagnostic pathways. Controversy also exists as to the optimum f-Hb to be used for referral for colonoscopy, although it is known that sensitivity will increase and specificity decrease as the cut-off f-Hb is lowered. Harmonisation of approaches to defining the detectability characteristics of FIT analytical methods is required as is standardisation of the reporting of results. FIT, with high sensitivity, provide a good rule in test for CRC and, with high NPV, a good rule out test for SBD: however, no test is perfect and, irrespective of the f-Hb cut-off applied, a small number of CRC will be missed as will rather more AA and IBD: thus, robust safety-netting is required for patients who have negative FIT results but continue to experience symptoms. Some evidence exists that a single sample of faeces is sufficient in assessment of patients with symptoms. Risk scoring models incorporating f-Hb and other variables, particularly age and sex, have been developed, but further research is required as to their value in clinical practice. Further simple research studies on the value of FIT in assessment of patients with symptoms would seem somewhat redundant. What is required now are reports on the evaluation of the routine use of FIT in assessment of patients with symptoms, particularly on the controversial aspects which still remain as outlined here, and the many other challenges not discussed here, but explicitly addressed in previous reviews, including many analytical aspects concerning the current analysis of f-Hb.[10,11,19]

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Conflicts of interest

CGF undertakes consultancy with Kyowa Medex Co., Ltd, Tokyo, Japan, and receives support to attend conferences from Alpha Labs Ltd, Eastleigh, Hants, UK.

References

1. Young GP, Symonds EL, Allison JE, Cole SR, Fraser CG, Halloran SP, et al. Advances in fecal occult blood tests: the FIT revolution. Dig Dis Sci. 2015;60:609–22, http://dx.doi.org/10.1007/s10620-014-3445-3.
2. Cram P, Fendrick AM, Inadomi J, Cowen ME, Carpenter D, Vijan S. The impact of a celebrity promotional campaign on the use of colon cancer screening: the Katie Couric effect. Arch Intern Med. 2003;163:1601–5, http://dx.doi.org/10.1001/archinte.163.13.1601.
3. Symonds EL, Fraser RJ, Young GP. FIT for purpose: enhanced applications for faecal immunochemical tests. J Lab Precis Med. 2018;3:28, http://dx.doi.org/10.21037/jlpm.2018.03.03.
4. Peacock O, Clayton S, Atkinson F, Tierney GM, Lund JN. Be Clear on Cancer: the impact of the UK National Bowel Cancer Awareness Campaign. Colorectal Dis. 2013;15:963–7, http://dx.doi.org/10.1111/codi.12220.
5. Jellema P, van der Windt DA, Bruinvels DJ, Mallen CD, van Weyenberg SJ, Mulder CJ, et al. Value of symptoms and additional diagnostic tests for colorectal cancer in primary care: systematic review and meta-analysis. BMJ. 2010;340:c1269, http://dx.doi.org/10.1136/bmj.c1269.
6. Vega P, Valentín F, Cubiella J. Colorectal cancer diagnosis: Pitfalls and opportunities. World J Gastrointest Oncol. 2015;7:422–33, http://dx.doi.org/10.4251/wjgo.v7.i12.422.
7. National Institute for Health and Care Excellence. NICE Guideline NG12; Suspected cancer: recognition and referral. July, 2017 (accessed 01 August 2018). https://www.nice.org.uk/guidance/ng12.
8. Fraser CG, Strachan JA. A nicer approach to the use of'faecal occult blood tests' in assessment of the symptomatic. Ann Clin Biochem. 2016;53:5–6, http://dx.doi.org/10.1177/0004563215612504.
9. Westwood M, Lang S, Armstrong N, van Turenhout S, Cubiella J, Stirk L, et al. Faecal immunochemical tests (FIT) can help to rule out colorectal cancer in patients presenting in primary care with lower abdominal symptoms: a systematic review conducted to inform new NICE DG30 diagnostic guidance. BMC Med. 2017;15:189, http://dx.doi.org/10.1186/s12916-017-0944-z.
10. Steele RJC, Fraser CG. Faecal immunochemical tests (FIT) for haemoglobin for timely assessment of patients with symptoms of colorectal disease. In: Olsson L Ed. Timely diagnosis of colorectal cancer. Cham: Springer; 2018, http://dx.doi.org/10.1007/978-3-319-65286-3_3.
11. Fraser CG. Faecal immunochemical tests (FIT) in the assessment of patients presenting with lower bowel symptoms: Concepts and challenges. Surgeon. 2018 Mar 13, http://dx.doi.org/10.1016/j.surge.2018.01.004, pii: S1479-666X(18)30019-2 [Epub ahead of print].
12. Juul JS, Hornung N, Andersen B, Laurberg S, Olesen F, Vedsted P. The value of using the faecal immunochemical test in general practice on patients presenting with non-alarm symptoms of colorectal cancer. Br J Cancer. 2018 Aug 1, http://dx.doi.org/10.1038/s41416-018-0178-7 [Epub ahead of print].
13. National Institute for Health and Care Excellence. NICE Diagnostic Guidance DG 30. July, 2017. Quantitative faecal immunochemical tests to guide referral for colorectal cancer in primary care. https://www.nice.org.uk/guidance/dg30. (accessed 01 August 2018).
14. Allison JE, Fraser CG, Halloran SP, Young GP. Population screening for colorectal cancer means getting FIT: the past, present, and future of colorectal cancer screening using the fecal immunochemical test for hemoglobin (FIT). Gut Liver. 2014;8:117–30, http://dx.doi.org/10.5009/gnl.2014.8.2.117.
15. Daly JM, Xu Y, Levy BT. Which fecal immunochemical test should I choose? J Prim Care Community Health. 2017;8:264–77, http://dx.doi.org/10.1177/2150131917705206.
16. Gies A, Cuk K, Schrotz-King P, Brenner H. Direct comparison of diagnostic performance of 9 quantitative fecal immunochemical tests for colorectal cancer screening. Gastroenterology. 2018;154:93–104, http://dx.doi.org/10.1053/j.gastro.2017.09.018.
17. Nicholson BD, Thompson M, Price CP, Heneghan C, Plüddemann A. Home-use faecal immunochemical testing: primary care diagnostic technology update. Br J Gen Pract. 2015;65:156–8, http://dx.doi.org/10.3399/bjgp15X684229.
18. Westwood M, Corro Ramos I, Lang S, Luyendijk M, Zaim R, et al. Faecal immunochemical tests to triage patients with lower abdominal symptoms for suspected colorectal cancer referrals in primary care: a systematic review and cost-effectiveness analysis. Health Technol Assess. 2017;21:1–234, http://dx.doi.org/10.3310/hta21330.
19. Godber IM, Benton SC, Fraser CG. Setting up a service for a faecal immunochemical test for haemoglobin (FIT): a review of considerations, challenges and constraints. J Clin Pathol 2018 Oct.1. pii:jclinpath-2018-205047, https://doi.org/10.1136/jclinpath-2018-0205047 [Epub ahead of print].
20. Gnatta E, Zaninotto M, Epifani MG, Padoan A, Gjini R, Plebani M. A new sampling device for faecal immunochemical testing: haemoglobin stability is still an open issue. Clin Chem Lab Med. 2014;52:1203–9, http://dx.doi.org/10.1515/cclm-2013-1074.
21. Grazzini G, Ventura L, Rubeca T, Rapi S, Cellai F, Di Dia PP, et al. Impact of a new sampling buffer on faecal haemoglobin stability in a colorectal cancer screening programme by the faecal immunochemical test. Eur J Cancer Prev. 2017;26:285–91, http://dx.doi.org/10.1097/CEJ. 0000000000000257.
22. Fraser CG. Comparison of quantitative faecal immunochemical tests for haemoglobin (FIT) for asymptomatic population screening. Transl Cancer Res. 2016;5 Suppl 4:S916–9, http://dx.doi.org/10.21037/tcr.2016.10.22.
23. Grobbee EJ, van der Vlugt M, van Vuuren AJ, Stroobants AK, Mundt MW, Spijker WJ, et al. A randomised comparison of two faecal immunochemical tests in population-based colorectal cancer screening. Gut. 2017;66:1975–82, http://dx.doi.org/10.1136/gutjnl-2016-311819.
24. Passamonti B, Malaspina M, Fraser CG, Tintori B, Carlani A, D'Angelo V, et al. A comparative effectiveness trial of two faecal immunochemical tests for haemoglobin (FIT). Assessment of test performance and adherence in a single round of a population-based screening programme for colorectal cancer. Gut. 2018;67:485–96, http://dx.doi.org/10.1136/gutjnl-2016-312716.
25. Wieten E, de Klerk CM, van der Steen A, Ramakers CR, Kuipers EJ, Hansen BE, et al. Equal accuracy of 2 quantitative fecal immunochemical tests in detecting advanced neoplasia in an organized colorectal cancer screening program. Gastroenterology. 2018 Jul 25, http://dx.doi.org/10.1053/

j.gastro.2018.07.021, pii: S0016-5085(18)34809-1[Epub ahead of print].
26. Auge JM, Rodriguez C, Espanyol O, Rivero L, Sandalinas S, Grau J, et al. An evaluation of the SENTiFIT 270 analyser for quantitation of faecal haemoglobin in the investigation of patients with suspected colorectal cancer. Clin Chem Lab Med. 2018;56:625–33, http://dx.doi.org/10.1515/cclm-2017-0605.
27. Mowat C, Digby J, Strachan JA, Wilson R, Carey FA, Fraser CG, et al. Faecal haemoglobin and faecal calprotectin as indicators of bowel disease in patients presenting to primary care with bowel symptoms. Gut. 2016;65:1463–9, http://dx.doi.org/10.1136/gutjnl-2015-309579.
28. 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, http://dx.doi.org/10.1373/clinchem.2011.177980.
29. Elias SG, Kok L, de Wit NJ, Witteman BJM, Goedhard G, Romberg-Camp MLJ, et al. Is there an added value of faecal calprotectin and haemoglobin in the diagnostic work-up for primary care patients suspected of significant colorectal disease? A cross-sectional diagnostic study. BMC Med. 2016;14:141, http://dx.doi.org/10.1186/s12916-016-0684-5.
30. Fraser CG. Diagnostic work-up of patients presenting in primary care with lower abdominal symptoms: which faecal test and triage strategy should be used? BMC Med. 2016;14:139, http://dx.doi.org/10.1186/s12916-016-0694-3.
31. Quyn AJ, Steele RJ, Digby J, Strachan JA, Mowat C, McDonald PJ, et al. Application of NICE guideline NG12 to the initial assessment of patients with lower gastrointestinal symptoms: not FIT for purpose? Ann Clin Biochem. 2018;55:69–76, http://dx.doi.org/10.1177/0004563217707981.
32. Högberg C, Karling P, Rutegård J, Lilja M. Diagnosing colorectal cancer and inflammatory bowel disease in primary care: The usefulness of tests for faecal haemoglobin, faecal calprotectin, anaemia and iron deficiency. A prospective study. Scand J Gastroenterol. 2017;52:69–75, http://dx.doi.org/10.1080/00365521.2016.1228120.
33. Ribbing Wilén H, Blom J, Höijer J, Hultcrantz R. Fecal immunochemical test in colorectal cancer screening: colonoscopy findings by different cut-off levels. J Gastroenterol Hepatol. 2018 Jul 3, http://dx.doi.org/10.1111/jgh.14373 [Epub ahead of print].
34. Rodríguez-Alonso L, Rodríguez-Moranta F, Ruiz-Cerulla A, Lobatón T, Arajol C, Binefa G, et al. An urgent referral strategy for symptomatic patients with suspected colorectal cancer based on a quantitative immunochemical faecal occult blood test. Dig Liver Dis. 2015;47:797–804, http://dx.doi.org/10.1016/j.dld.2015.05.004. 7.
35. Cubiella J, Salve M, Díaz-Ondina M, Vega P, Alves MT, Iglesias F, et al. Diagnostic accuracy of the faecal immunochemical test for colorectal cancer in symptomatic patients: comparison with NICE and SIGN referral criteria. Colorectal Dis. 2014;16:O273–82, http://dx.doi.org/10.1111/codi.12569.
36. McDonald PJ, Digby J, Innes C, Strachan JA, Carey FA, Steele RJ, et al. Low faecal haemoglobin concentration potentially rules out significant colorectal disease. Colorectal Dis. 2013;15:e151–9, http://dx.doi.org/10.1111/codi.12087.
37. Godber IM, Todd LM, Fraser CG, MacDonald LR, Younes HB. Use of a faecal immunochemical test for haemoglobin can aid in the investigation of patients with lower abdominal symptoms. Clin Chem Lab Med. 2016;54:595–602, http://dx.doi.org/10.1515/cclm-2015-0617.
38. Widlak MM, Thomas CL, Thomas MG, Tomkins C, Smith S, O'Connell N, et al. Diagnostic accuracy of faecal biomarkers in detecting colorectal cancer and adenoma in symptomatic patients. Aliment Pharmacol Ther. 2017;45:354–63, http://dx.doi.org/10.1111/apt.13865.
39. Fraser CG. Interpretation of faecal haemoglobin concentration data in colorectal cancer screening and in assessment of symptomatic patients. J Lab Precis Med. 2017;2:96, http://dx.doi.org/10.21037/jlpm.2017.11.01.
40. Fraser CG, Benton SC. Detection capability of quantitative faecal immunochemical tests for haemoglobin (FIT) and reporting of low faecal haemoglobin concentrations. Clin Chem Lab Med. 2018 Jul 11, http://dx.doi.org/10.1515/cclm-2018-0464 [Epub ahead of print] pii:/j/cclm.ahead-of-print/cclm-2018-0464/cclm-2018-0464.xml.
41. Spiteri N, Skaife P, Get FIT. for the new year: a review of the role of faecal immunochemical test for haemoglobin in patients with symptoms of colorectal disease. J Lab Precis Med. 2018;3:52, http://dx.doi.org/10.21037/jlpm.2018.05.05.
42. Högberg C, Söderström L, Lilja M. Faecal immunochemical tests for the diagnosis of symptomatic colorectal cancer in primary care: the benefit of more than one sample. Scand J Prim Health Care. 2017;35:369–72, http://dx.doi.org/10.1080/02813432.2017.1397255.
43. Auge JM, Fraser CG, Rodriguez C, Roset A, Lopez-Ceron M, Grau J, et al. Clinical utility of one versus two faecal immunochemical test samples in the detection of advanced colorectal neoplasia in symptomatic patients. Clin Chem Lab Med. 2016;54:125–32, http://dx.doi.org/10.1515/cclm-2015-0388.
44. Williams TG, Cubiella J, Griffin SJ, Walter FM, Usher-Smith JA. Risk prediction models for colorectal cancer in people with symptoms: a systematic review. BMC Gastroenterol. 2016;16:63, http://dx.doi.org/10.1186/s12876-016-0475-7.
45. McDonald PJ, Strachan JA, Digby J, Steele RJ, Fraser CG. Faecal haemoglobin concentrations by gender and age: implications for population-based screening for colorectal cancer. Clin Chem Lab Med. 2011;50:935–40, http://dx.doi.org/10.1515/CCLM.2011815.
46. Fraser CG, Rubeca T, Rapi S, Chen LS, Chen HH. Faecal haemoglobin concentrations vary with sex and age, but data are not transferable across geography for colorectal cancer screening. Clin Chem Lab Med. 2014;52:1211–6, http://dx.doi.org/10.1515/cclm-2014-0115.
47. Cubiella J, Vega P, Salve M, Díaz-Ondina M, Alves MT, Quintero E, et al. Development and external validation of a faecal immunochemical test-based prediction model for colorectal cancer detection in symptomatic patients. BMC Med. 2016;14:128, http://dx.doi.org/10.1186/s12916-016-0668-5.
48. Cubiella J, Digby J, Rodríguez-Alonso L, Vega P, Salve M, Díaz-Ondina M, et al. The fecal hemoglobin concentration, age and sex test score: development and external validation of a simple prediction tool for colorectal cancer detection in symptomatic patients. Int J Cancer. 2017;140:2201–11, http://dx.doi.org/10.1002/ijc.30639.
49. Digby J, McDonald PJ, Strachan JA, Libby G, Steele RJ, Fraser CG. Deprivation and faecal haemoglobin: implications for bowel cancer screening. J Med Screen. 2014;21:95–7, http://dx.doi.org/10.1177/0969141314535388.
50. Symonds EL, Osborne JM, Cole SR, Bampton PA, Fraser RJ, Young GP. Factors affecting faecal immunochemical test positive rates: demographic, pathological, behavioural and environmental variables. J Med Screen. 2015;22:187–93, http://dx.doi.org/10.1177/0969141315584783.
51. Senore C, Haug U. Faecal immunochemical tests have the potential for correctly ruling out colorectal cancer in symptomatic patients. BMJ Evid Based Med. 2018;23:113–4, http://dx.doi.org/10.1136/bmjebm-2018-110901.