Interval cancers using a quantitative faecal immunochemical test (FIT) for haemoglobin when colonoscopy capacity is limited

Digby, Jayne; Fraser, Callum G.; Carey, Francis A.; Lang, Jaroslaw; Stanners, Greig; Steele, Robert

Published in:
Journal of Medical Screening

DOI:
10.1177/0969141315609634

Publication date:
2016

Document Version
Peer reviewed version

Interval cancers using a quantitative faecal immunochemical test for haemoglobin (FIT) when colonoscopy capacity is limited.

Jayne Digby,[1,2] Callum G Fraser,[1,2] Francis A Carey,[3] Jaroslaw Lang,[4] Greig Stanners, [4] Robert JC Steele.[1,2,5]

[1]Scottish Bowel Screening Research Unit, Ninewells Hospital and Medical School, Dundee, Scotland

[2]Centre for Research into Cancer Prevention and Screening, Ninewells Hospital and Medical School, Dundee, Scotland

[3]Department of Pathology, Ninewells Hospital and Medical School, Dundee, Scotland

[4]Information Services Division, NHS National Services Scotland, Meridian Court, Glasgow, Scotland

[5]Medical Research Institute, Division of Cancer, Ninewells Hospital and Medical School, Scotland

Correspondence to: Jayne Digby, Scottish Bowel Screening Research Unit**,** Mailbox 7, Level 7, Ninewells Hospital and Medical School, Dundee DD1 9SY, Scotland: jaynedigby@nhs.net

Tel: 01382383982

Short Title: Interval cancers when colonoscopy is limited.

Keywords: colorectal cancer: faecal haemoglobin: faecal immunochemical test: faecal occult blood test; interval cancers:

Number of words (abstract): 250

Number of words (text): 2567

Number of Tables: 3

Number of Figures: 1

Number of references: 26

Abstract

Objectives Quantitative faecal immunochemical tests for haemoglobin (FIT) in colorectal cancer (CRC) screening pose challenges when colonoscopy is limited. For low positivity rates, high faecal haemoglobin concentration (f-Hb) cut-offs are required, but little is known about interval cancer (IC) proportions using FIT. We assessed IC proportions using an 80 μg Hb/g cut-off.

Setting Two NHS Boards in the Scottish Bowel Screening Programme in which evaluation of FIT as a first-line test was performed.

Methods f-Hb was estimated for 30893 participants aged 50-75 years: 753 participants with f-Hb ≥ 80 μg Hb/g were referred for colonoscopy. IC, defined as CRC within two years of a negative result, were identified from the Scottish Cancer Registry.

Results There were 31 IC and 30 screen-detected (SD) CRCs, an IC proportion of 50.8%: 48.4%: for men and 53.3%: for women. CRC site distribution was similar between IC and SD, but IC were later stage (46.7% and 33.3%: Dukes' stages C and D, respectively). Of 31 IC, 23 had f-Hb <10 μg Hb/g including six with undetectable f-Hb. A f-Hb cut-off of 10 μg Hb/g would have raised the positivity rate from 2.4% to 9.4%, increased colonoscopy requirement from 753 to 2147, and reduced the IC proportion to 38.3%.

Conclusions The IC proportion was similar to that seen with guaiac-based FOBT. The later stage distribution of IC highlights the benefits of lower f-Hb cut-offs, but with 19.4% of IC having undetectable f-Hb, some cancers would have been missed, even with drastic reduction in the f-Hb cut-off.

Introduction

Interval cancers (IC) are a significant issue in colorectal cancer (CRC) screening programmes. The Expert Working Group for Right-Sided Lesions and Interval Cancers, Colorectal Cancer Screening Committee, World Endoscopy Organization, recently recommended a standardised nomenclature for IC across all CRC screening modalities and colonoscopy surveillance, providing a definition of IC as: CRC diagnosed after a screening test or examination in which no cancer is detected and before the date of the next recommended examination.[1] Minimising the number of undetected CRC is crucial to the success of CRC screening programmes in meeting their primary goal of reducing CRC mortality through early detection.

Although randomised controlled trials (RCTs) have shown that guaiac faecal occult blood test (gFOBT) screening reduces CRC mortality,[2] high proportions of all CRC diagnosed in the screened population that were IC, here referred to as the IC proportion, are commonly reported. In England[3] and Denmark,[4] IC proportions of 51.3% and 55.2% were found, and a large non-randomised trial in Burgundy calculated the IC proportion at 59.3%.[5] Further studies from Denmark[6] Scotland,[7] and France[8] have also provided evidence that IC consistently account for more than half of CRC detected in populations screened biennially with gFOBT.

Previously published work has also identified some characteristics more associated with IC than gFOBT screen-detected (SD) CRC. Higher proportions of IC are found in women compared with SD CRC[7,9,10] and significantly more IC have been demonstrated to arise in the right colon than SD CRC.[7,9–15] Assuming advanced neoplasia was present at the time of the negative gFOBT, these findings suggest that

gFOBT may be more likely to detect pathology in men and in the left side of the colon. Furthermore, rectal cancers have been found to be more common amongst IC than SD CRC, [6–8] perhaps because tumour growth is faster for rectal cancer,[16] or because the erythrocytes in any blood originating in the rectum have not been haemolysed and the still intact erythrocytes do not yield positive gFOBT or FIT. IC have also been associated with a worse prognosis, with larger, later stage tumours more frequently reported for IC compared with SD CRC. [6–8]

Countries worldwide are now introducing faecal immunochemical tests for haemoglobin (FIT) to replace gFOBT in CRC screening programmes, in view of their many advantages[17] With numerous studies demonstrating FIT to be more sensitive tests than gFOBT, particularly for advanced adenoma detection,[18] it is likely that FIT have the potential to reduce IC proportions. Moreover, quantitative FIT allow programme organisers to select a faecal haemoglobin concentration (f-Hb) cut-off most appropriate for their programme. However, this poses considerable challenges for countries with limited colonoscopy capacity. To secure low positivity rates that match colonoscopy capacity, high f-Hb cut-offs must be used, negating the improved sensitivity of FIT over gFOBT; this has been demonstrated by the results of an evaluation of quantitative FIT in Scotland at a cut-off f-Hb of 80 µg Hb/g faeces in which the positive predictive values (PPV) for advanced neoplasia were no better than with gFOBT.[19]

Thus, it is important to establish the IC proportions associated with the use of FIT at a cut-off equivalent to gFOBT and if characteristics such as female sex and location in the proximal colon continue to show positive associations with IC. However, data on IC proportions in population screening with FIT are lacking; these would provide essential insights into how quantitative FIT can be utilised in countries with limited colonoscopy capacity to minimise IC proportions and address the sex inequalities

that exist with gFOBT screening. In consequence, we assessed the consequences of FIT using a f-Hb cut-off of 80 µg Hb/g faeces (set to give ca. 2% positivity) in terms of IC within an established CRC screening programme.

Methods

The FIT as a first-line test evaluation has been described previously.[19] From 01 July, 2010, to 12 January, 2011, all eligible participants in the Scottish Bowel Screening Programme resident in NHS Tayside and NHS Ayrshire & Arran were sent a FIT kit pack containing an invitation letter, a booklet on bowel cancer, a thin card wallet with written and pictorial instructions for sample collection which contained a single faecal specimen collection device (Eiken Chemical Co. Ltd., Tokyo, Japan), a small zip-lock plastic bag with integral absorbent material, and a foil mailing pouch for device return. The population invited for screening were individuals aged 50–74 years. Both NHS Boards had offered screening previously, using a gFOBT/qualitative FIT two-tier reflex screening algorithm,[20] but this was the first time that any participant had taken part in a quantitative FIT-based CRC screening programme. Those who sent an untestable FIT were sent another FIT kit pack.

The characteristics of all returned samples were documented and the samples analysed for f-Hb using OC-Sensor Diana automated immunoturbidimetric analysers (Eiken). Analyses were carried out in the Scottish Bowel Screening Centre Laboratory by trained staff whose major function is to perform faecal test analyses; the Laboratory has a comprehensive total quality management system and is accredited to ISO15189 based standards by Clinical Pathology Accreditation (UK) Ltd.

All participants with f-Hb < 80 µg Hb/g faeces were reported as negative and

informed by letter. All participants with f-Hb $\geq$ 80 µg Hb/g faeces were reported as positive and contacted by letter, the general practitioner was notified, and the individual was referred to their NHS Board for colonoscopy. The f-Hb cut-off was chosen to give ca. 2% positivity, to mimic the positivity rate of the existing Screening Programme and match the available colonoscopy resource. Data for colonoscopy outcomes and any subsequent pathology were downloaded from the appropriate NHS Tayside and NHS Ayrshire & Arran clinical IT systems. Data on colonoscopy findings, including number, size, Dukes' stage, and localisation of colorectal cancers and adenomas were collected. Right-sided location of neoplasia was defined as cancer detected in the region of the colon up to and including the splenic flexure, left-sided as the region thereafter up to the recto-sigmoid junction, and rectal neoplasia as lesions located both in the recto-sigmoid junction and the rectum.

Linkage with the Scottish Cancer Registry was performed to identify IC from the cohort of negative participants and to allow comparison of factors including f-Hb and gender distribution of CRC between the group with IC and those with SD CRC. IC data was available for IC diagnosed up to 31 December 2012, meaning that the analysis included only negative participants with a result date up to 31 December 2010 and participants with a later result date were excluded from the analysis. The linkage was completed using *IBM SPSS* version 21. CRC arising after a negative colonoscopy were referred to as "missed" cancers, and not IC in our cohort.

Population weighted Scottish Index of Multiple Deprivation (SIMD) 2012 quintiles were used for analysis by deprivation.[21]

MedCalc (MedCalc Software, Mariakerke, Belgium) statistical software was used for all calculations. The Mann-Whitney U test was used for comparison of median f-Hb between groups. Probability of $p < 0.05$ was considered significant. Logistic

regression analysis was performed to calculate odds ratios (OR) for IC amongst different demographic groups, adjusted for confounding variables.

Results

Over the six month screening period for which IC data was available, a total of 30893 participants in the two NHS Boards responded to screening, with 30140 participants negative and 753 having a f-Hb above the 80 µg Hb/g cut-off concentration for positivity. 104 positive participants did not complete follow-up investigations due to either non-attendance, recently performed colonoscopy, or being deemed unfit for invasive procedures, and were excluded from further analysis. Of 649 participants completing investigations as a result of their positive FIT result, 30 had SD CRC. 31 cases of IC were identified from follow-up of participants with a negative screening result to give an IC proportion of 50.8%. Table 1 shows the characteristics associated with IC and SD.

In men, 48.4% of all CRC were IC and, in women, 50.3% were IC. Median age in those with an IC was 68 years (interquartile range [IQR]: 63-72) compared with 67 years (interquartile range: 61-72) for SD CRC cases. IC were diagnosed at a more advanced stage than SD CRC with 46.7% of IC being late stage (Dukes' stage C or D) compared with 33.3% of SD CRC.

Table 1. Characteristics of interval cancers and screen-detected colorectal cancer.

	Interval cancers		Screen-detected		p-value
	n	%	n	%	
Total cases	31	50.8	30	49.2	1.00
Sex					
Men	15	48.4	16	51.6	0.90
Women	16	53.3	14	46.7	

Age quintile (years)*					
50-54	0	0.0	4	100.0	
55-59	3	50.0	3	50.0	
60-64	8	66.7	4	33.3	0.22
65-69	5	38.5	8	61.5	
70-74	15	57.7	11	42.3	
Deprivation (SIMD)					
1 (most deprived)	5	50.0	5	50.0	
2	4	66.7	2	33.3	
3	5	35.7	9	64.3	0.95
4	9	60.0	6	40.0	
5 (least deprived)	8	50.0	8	50.0	
Cancer site**					
Right-sided	13	50.0	13	50.0	
Left-sided	5	50.0	5	50.0	0.99
Rectum	13	52.0	12	48.0	
Dukes' stage					
A	6	42.9	8	57.1	
B	10	50.0	10	50.0	0.07
C	7	43.8	9	56.3	
D	7	100.0	0	0.0	
Not known	1	25.0	3	75.0	

* age at time of invite.

** right-sided CRC includes region up to and including the splenic flexure; left-sided includes descending and sigmoid colon; rectum includes recto-sigmoid junction and rectum.

Table 2 shows median f-Hb and corresponding IQR at the time of screening in those who were subsequently found to have an IC, allowing comparison of f-Hb according to sex, CRC stage, site and time to diagnosis following screening.

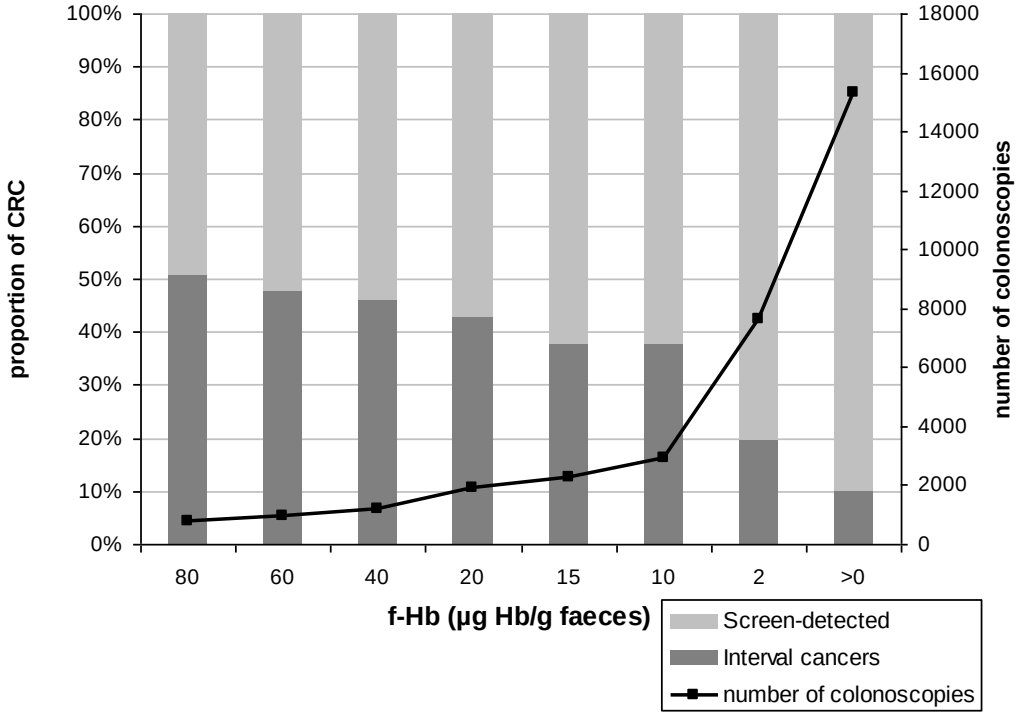
Table 2. Median f-Hb and interquartile range at time of negative screening test in those who had interval cancer.

	n	Median f-Hb (µg Hb/g faeces)	IQR	p-value
All	31	2.8	0.4-13.5	
Sex				
Men	15	12.6	0.1-12.9	0.44
Women	16	0.5	0.4-11.7	
Stage				
Total early	16	3.1	0.4-12.1	0.47
Total late	14	2.5	0.4-6.8	
Site				
Proximal	13	1.4	0.0-11.8	0.39
Distal	18	3.1	0.4-15.2	
Time to diagnosis				

within 1 year	8	4.1	0.2-7.5	0.79
1-2 years	23	2.8	0.4-15.9	

The effect on positivity rate and IC proportion of lowering the f-Hb cut-off to various concentrations was assessed. Between cut-off concentrations of 80 µg Hb/g faeces and 10 µg Hb/g faeces, positivity rate would have increased steadily from 2.4% to 9.4%, before escalating to 24.7% at 2 µg Hb/g faeces and 49.5% using a cut-off concentration of any detectable blood. Figure 1 shows the number of colonoscopies that would have been required at different f-Hb cut-offs alongside the associated proportions of IC and SD CRC.

Figure 1. Effect of lowering the faecal haemoglobin (f-Hb) cut-off on proportions of interval cancer and screen-detected colorectal cancers (CRC) and number of colonoscopies required.



Of the 31 IC cases, 23 had f-Hb < 10 µg Hb/g faeces at the time of their negative screening test, meaning that over a third of CRC cases would still have been missed if this f-Hb cut-off had been adopted. Furthermore, six of these 23 cases had undetectable f-Hb. With 51.8% of all participants in our evaluation having undetectable f-Hb,[19] the proportion of IC arising in this group was compared with those with higher f-Hb and produced OR for IC, using those with no detectable f-Hb as the reference. OR were also calculated for men compared with women and for those between 60-69 years and over 70 years compared with those aged 50-59 years. These results, along with OR also adjusted for all other factors investigated are displayed in Table 3.

Table 3. Proportion of interval cancers by faecal haemoglobin (f-Hb), gender and age with adjusted odds ratios.

	% with interval cancer	Adjusted odds ratio (95% CI)
f-Hb (µg Hb/g faeces)		
0	0.04	1.00
>0	0.17	3.84 (1.57 – 9.40)
>10	0.38	8.01 (2.73 – 23.56)
>20	0.44	8.74 (2.61 – 29.21)
>40	0.75	15.56 (3.76 – 64.33)
60-79.9	1.31	23.91 (4.73 – 120.81)
Sex		
Female	0.10	1.00
Male	0.11	1.01 (0.50 – 2.04)
Age (years)		
50-59	0.02	1.00
60-69	0.11	4.69 (1.33 – 16.47)
70+	0.32	12.16 (3.50 – 42.26)

Discussion

Our results provide unique insights into IC proportions using FIT with a high f-Hb cut-off (80 µg Hb/g faeces) in an established screening programme with limited

colonoscopy capacity and how these proportions could be influenced by varying the f-Hb cut-off. Our IC proportion was no different to the ca. 50% found using traditional gFOBT.

A major strength of this study is that a large cohort of over 30000 completed FIT screening in the context of a fully rolled out operational screening programme, meaning that the implications on IC proportions of using a FIT at a high f-Hb cut-off concentration are what could be expected on nationwide implementation of FIT screening. Previous reports on this topic are very limited, therefore our results provide important evidence that FIT-based CRC screening programmes would benefit from use of low f-Hb cut-offs to gain lower IC proportions as well as higher sensitivity and detection of earlier stage disease, but at the cost of increased colonoscopy demand.[22] This study also has limitations. The relatively small numbers of IC and SD CRC mean that statistical significance was not reached for differences between CRC categories for Dukes' stage, for example. It is also important to bear in mind that the distributions of f-Hb are country specific.[23] As a result, our findings may not necessarily be transferable. Screening programmes in other countries should perform their own analyses of IC proportions using FIT to determine the consequences of the use of different f-Hb cut-offs on IC. Moreover, our calculation of the yield of SD CRC at different f-Hb cut-offs is likely to be an underestimation. In addition to avoided IC, a lower f-Hb cut-off may also have led to detection of CRC that would arise as SD CRC the subsequent screening round as well as a small proportion of over-diagnosed cancers. Therefore, in reality the IC proportions would be lower than we have reported, although this is difficult to quantify.

Our IC proportion of 50.8% at a cut-off concentration of 80 µg Hb/g was much higher than the overall 14.4% IC proportion found in Italy with a much lower f-Hb cut-off of 20 µg Hb/g faeces,[24] confirming that the use of a high f-Hb cut-off negates the

improved sensitivity for significant neoplasia offered by FIT over gFOBT and consequently increases the IC proportion.[19] Previous findings that women have a higher IC proportion than men, at least when a high f-Hb cut-off is used, are supported to some extent. In attempting to explain this, we analysed characteristics associated with CRC between the sexes. In contrast to results of previous studies investigating IC in gFOBT screening programmes, an association with location in the proximal colon for IC in women was lacking, with just a quarter of cases in women located from the caecum up to and including the splenic flexure, whereas most IC in men were right-sided. However, the relatively small numbers of IC and SD CRC detected in this study make this lack of association unreliable, and indeed, the relationships between IC proportions and sex and age did not reach statistical significance although, reassuringly, the trends were as expected.

The more advanced stage distribution of IC highlights the need for measures to be taken to improve CRC detection with screening. Lowering the f-Hb cut-off would be an obvious solution, but the resultant increase in colonoscopy demand may not be sustainable given the available resources. For example, in our cohort, halving the f-Hb cut-off to 40 µg Hb/g faeces would reduce the IC proportion from 50.8% to only 45.9%, but with a significant 58.6% increase in the number of colonoscopies required. Small gains in sensitivity would come at the cost of significant losses in specificity and PPV. This problem could be counteracted by screening at a low f-Hb cut-off, but with a longer interval between screening rounds than the two years currently implemented, and investigating the impact of such a strategy on IC proportions is an important area for future research. In addition, given the important effects of age and gender on IC proportions seen in previous studies and supported here, exploring stratified f-Hb cut-offs based on these variables is warranted.

Although participants who had an undetectable f-Hb accounted for over half of the

screened population, the proportion of IC in this group was relatively small, over 30 times lower than the proportion of IC identified in those with f-Hb in the range 60.0 – 79.9 µg Hb/g faeces, who constituted just 0.5% of the cohort. Adjusted OR demonstrated increasing risk of IC with increasing f-Hb and perhaps indicate a need for participants with elevated f-Hb to be offered more regular screening. As we have suggested previously, since men and women have different f-Hb as do older participants compared to younger,[22,25] our results support the inclusion of numerical data for f-Hb in risk-scoring models for population CRC screening such as recently advocated.[26] With IC being associated with a worse prognosis, it appears that women and older participants may be disadvantaged by the use of a single f-Hb cut-off for all and we therefore consider that better individualised use of FIT in CRC screening is required. However, further work is required to determine specific performance characteristics of FIT in subgroups at different f-Hb cut-off concentrations. IC proportion alone is not sufficient to address inequalities; for instance CRC incidence rates are lower among women than men meaning that the residual risk of IC diagnosis following a negative screening result will also be lower in women than in men. Therefore, the selection of the appropriate f-Hb cut-off concentration is complex and other factors such as PPV are an important consideration in a setting limited by endoscopic resources.

Declaration of interests

CGF undertook consultancy with Immunostics Inc., Ocean, NJ, USA, Mode Diagnostics, Glasgow, Scotland, and Kyowa-Medex, Tokyo, Japan. All other authors had no conflicting interests.

Funding

This work was supported by The Scottish Government Health Directorates.

Research Ethics

Approval from the NHS National Services Scotland: Privacy Advisory Committee was also secured for the data linkage required in this analysis.

References

1. Sanduleanu S, le Clercq CM, Dekker E, et al. On behalf of the Expert Working Group on 'Right-sided lesions and interval cancers', Colorectal Cancer Screening Committee, World Endoscopy Organization.Definition and taxonomy of interval colorectal cancers: a proposal for standardising nomenclature. *Gut* 2014 Sep 5.[Epub ahead of print].

2. Towler B, Irwig L, Glasziou P, et al. A systematic review of the effects of screening for colorectal cancer using the faecal occult blood test, Hemoccult. *Br Med J* 1998;**317**:559– 65.

3. Hardcastle JD, Chamberlain JO, Robinson MHE, et al. Randomised controlled trial of faecal-occult-blood screening for colorectal cancer. *Lancet* 1996;**348**:1472– 7.

4. Kronborg O, Fenger C, Olsen J, Jorgensen OD, Sondergaard O. Randomised study of screening for colorectal cancer with faecal-occult-blood test. *Lancet* 1996;**348**:1467– 71.

5. Faivre J, Arveux P, Milan C, Durand G, Lamour J, Bedenne L. Participation in mass screening for colorectal cancer: results of screening and rescreening from the Burgundy study. *Eur J Cancer Prev* 1991;**1**:49– 55.

6. Jensen BM, Kronborg O, Fenger C. Interval cancers in screening with fecal occult blood test for colorectal cancer. *Scand J Gastroenterol* 1992;**27**:779–82.

7. Steele RJ, McClements P, Watling C, et al. Interval cancers in a FOBT-based colorectal cancer population screening programme: implications for stage, gender and tumour site. *Gut* 2012;**61**:576– 81.

8. Tazi MA, Faivre J, Lejeune C, Bolard P, Phelip JM, Benhamiche AM. Interval cancers in a community-based programme of colorectal cancer screening with faecal occult blood test. *Eur J Cancer Prev* 1999;**8**:13– 15.

9. Gill MD, Bramble MG, Rees CJ, Lee TJ, Bradburn DM, Mills SJ. Comparison of screen-detected and interval colorectal cancers in the Bowel Cancer Screening Programme. *Br J Cancer* 2012;**107**:417– 21.

10. Brenner H, Chang-Claude J, Seiler CM, Hoffmeister M. Interval cancers after negative colonoscopy: population-based case-control study. *Gut* 2012;**61**:15768–2.

11. Cooper GS, Xu F, Barnholtz Sloan JS, Schluchter MD, Koroukian SM. Prevalence and predictors of interval colorectal cancers in Medicare beneficiaries. *Cancer* 2012;**118**:3044– 52.

12. Singh H, Turner D, Xue L, et al. Risk of developing colorectal cancer following a negative colonoscopy examination: evidence for a 10-year interval between colonoscopies. *JAMA* 2006;**295**:2366– 73.

13. Singh H, Nugent Z, Demers AA, et al. Rate and predictors of early/missed colorectal cancers after colonoscopy in Manitoba: a population-based study. Am J *Gastroenterol* 2010;**105**:2588– 96.

14. Farrar WD, Sawhney MS, Nelson DB, et al. Colorectal cancers found after a complete colonoscopy. *Clin Gastroenterol Hepatol* 2006;**4**:1259– 64.

15. Hosokawa O, Shirasaki S, Kaizaki Y, et al. Invasive colorectal cancer detected up to 3 years after a colonoscopy negative for cancer. *Endoscopy* 2003;**35**:506–10.

16. Launoy G, Smith TC, Duffy SW , Bouvier V. Colorectal cancer mass-screening: Estimation of faecal occult blood test sensitivity, taking into account cancer mean sojourn time. *Int J Cancer* 1997:**73:**220–4.

17. 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.

18. Rabeneck L, Rumble RB, Thompson F, et al. Fecal immunochemical tests compared with guaiac fecal occult blood tests for population-based colorectal cancer screening. *Can J Gastroenterol* 2012;**26**:131-47.

19. Steele RJC, McDonald PJ, Digby J, et al. Clinical outcomes using a faecal immunochemical test for haemoglobin as a first-line test in a national programme constrained by colonoscopy capacity. *United European Gastroenterology J* 2013;**1:**198– 205.

20. Fraser CG, Digby J, McDonald PJ, Strachan JA, Carey FA, Steele RJ. Experience with a two-tier reflex gFOBT/FIT strategy in a national bowel

screening programme. *J Med Screen* 2012;**19**:8-13.

21. The Information Services Division. The Scottish Index of Multiple Deprivation (SIMD). Available from: http://www.isdscotland.org/Products-and-Services/GPD-Support/Deprivation/SIMD/ (accessed 16th March 2015).

22. Young GP, Symonds EL, Allison JE, et al. Advances in fecal occult blood tests: The FIT revolution. *Dig Dis Sci* 2015;**60**:609-22.

23. 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.

24. Zorzi M, Fedato C, Grazzini G, et al. High sensitivity of five colorectal screening programmes with faecal immunochemical test in the Veneto Region, Italy. *Gut* 2011;**60**:944–9.

25. 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**:9354–0.

26. Auge JM, Pellise M, Escudero JM, et al. Risk stratification for advanced colorectal neoplasia according to fecal hemoglobin concentration in a colorectal cancer screening program. *Gastroenterol* 2014;**147**:628-36.