Risk profiling and screening for colorectal cancer
Stegeman, I.

Citation for published version (APA):

General rights
It is not permitted to download or to forward/distribute the text or part of it without the consent of the author(s) and/or copyright holder(s), other than for strictly personal, individual use, unless the work is under an open content license (like Creative Commons).

Disclaimer/Complaints regulations
If you believe that digital publication of certain material infringes any of your rights or (privacy) interests, please let the Library know, stating your reasons. In case of a legitimate complaint, the Library will make the material inaccessible and/or remove it from the website. Please Ask the Library: http://uba.uva.nl/en/contact, or a letter to: Library of the University of Amsterdam, Secretariat, Singel 425, 1012 WP Amsterdam, The Netherlands. You will be contacted as soon as possible.
Risk factors for false positive and for false negative test results in screening with Fecal Occult Blood testing


*International Journal of Cancer; 2013; Epub ahead of print*
Abstract

Differences in the risk of a false negative or a false positive Fecal immunochemical Test (FIT) across subgroups may affect optimal screening strategies. We evaluate whether subgroups are at increased risk of a false positive or a false negative FIT result, whether such variability in risk is related to differences in FIT sensitivity and specificity or to differences in prior CRC risk.

Randomly selected, asymptomatic individuals were invited to undergo colonoscopy. Participants were asked to undergo one sample FIT and to complete a risk questionnaire. We identified patient characteristics associated with a false negative and false positive FIT results using logistic regression. We focused on statistically significant differences as well as on variables influencing the false positive or negative risk for which the odds ratio exceeded 1.25.

Of the 1,426 screening participants, 1,112 (78%) completed FIT and the questionnaire; 101 (9.1%) had advanced neoplasia. 102 Individuals were FIT positive, 65 (64%) had a false negative FIT result and 66 (65%) a false positive FIT result. Participants at higher age and smokers had a significantly higher risk of a false negative FIT result. Males were at increased risk of a false positive result, so were smokers and regular NSAID users. FIT sensitivity was lower in females. Specificity was lower for males, smokers and regular NSAID users. FIT sensitivity was lower in women. FIT specificity was lower in males, smokers and regular NSAID users. Our results can be used for further evidence based individualization of screening strategies.
Background

Colorectal cancer (CRC) is one of the leading causes of cancer related death. Detecting cancer or one of its precursors at an early stage can prevent premature death and may reduce cancer morbidity, since treatment for earlier-stage cancers is less aggressive than that for more advanced stage cancers. The high incidence of CRC, the high burden of disease, the availability of screening tests and of effective treatment of adenomas make CRC a likely candidate for population screening.

Colonoscopy is the reference standard for the detection of advanced adenoma and cancer in CRC screening. As it is a burdensome and costly procedure, fecal immunochemical stool testing (FIT) is often used as a non-invasive triaging test for colonoscopy in colorectal cancer screening programs. Given its considerable false negative rate in the detection of advanced neoplasia, FIT should be applied at regular intervals for an optimal preventive effect. With a specificity of 94% at the 50ng/ml cut off, the accuracy of FIT based screening is sub-optimal. A false positive test result will lead to unnecessary colonoscopies, accompanied by extra costs, burden and risks for the patient, which could affect participation in screening programs. It has been shown that women with a false positive mammography were less likely to attend to subsequent screening rounds. Also, being recalled for further diagnostic tests can evoke high levels of (in the case of a false positive test, unnecessary) anxiety and a lower quality of live.

It is possible that the risk of having a false positive or false negative FIT result varies across subgroups. In that case, the screening strategy could be adjusted for specific patient profiles in order to improve the effectiveness and efficiency of a screening program. Tailored screening strategies for patients at several risk levels could be introduced, in which the screening frequency is adjusted according to baseline CRC risk. This approach can only be justified if test characteristics are stable across risk groups.

So far, the evidence about risk factors for false positivity and differences in FIT accuracy in CRC screening is limited. According to Chiang et al individuals using antiplatelet drugs have an increased risk of having a false positive FIT. To strengthen the evidence base for developing CRC screening programs, we analyzed data from a population screening study where all participants completed a risk questionnaire, submitted a FIT and underwent a full colonoscopy.
Chapter 7  |  Risk factors for false positive and for false negative test results in screening with Fecal Occult Blood testing

Methods

Study population

Data were collected in the Colonoscopy or Colonography for Screening (COCOS) study, a multicenter population-based CRC screening pilot in two academic centers in the Netherlands. At the time of the study, the Netherlands had no nationwide CRC screening program. In the COCOS trial participation and yield in a population based CRC screening program were compared between colonoscopy and CT colonography as primary screening methods. The trial is described in detail elsewhere, a summary is given below.7,8

For our analyses we only included data from participants invited for primary colonoscopy. These were 6,600 randomly selected and asymptomatic men and women between 50 and 75 years of age. Invitees who had a full colonic examination in the previous 5 years were excluded from the program, as well as those who were in a colonoscopy surveillance program and people with a life-expectancy below 5 years.

FIT

All screening participants were invited to perform a one sample FIT (OC- Sensor, Eiken Chemical Co., LTD., Japan) prior to their colonoscopy. They were verbally instructed, during a pre-assessment appointment at the screening center, and received written instructions. Participants were instructed to perform the FIT at home within 48 hours before colonoscopy, but before starting the bowel preparation and were asked to take the sample to the screening center. Another option for FIT collection was to call the screening center immediately after performing the FIT, if this option was chosen the FIT was collected by research staff within 48 hours at the home of the participant. All FITs were stored at a -20 degree Celsius freezer upon collection at the screening center and automatically analyzed within 6 weeks after collection. Detailed information about the FIT testing and results is described elsewhere.3

Colonoscopy

All colonoscopies were performed at one of the two screening centers by gastroenterologists with an experience of more than 1,000 colonoscopies. Colonoscopies were done using the standard quality aspects defined by the American Society for Gastrointestinal Endoscopy Gastrointestinal Endoscopy.9 Participants were prepared for colonoscopy by a low fiber diet, 2 L of hypertonic polyethylene glycol solution (Moviprep; Norgine bv, Amsterdam, The Netherlands) and 2 L of fluids. The procedure was performed under conscious sedation using intravenous midazolam (Dormicum, Actavis, Baarn, The Netherlands) and fentanyl (Bipharma, Weesp, The Netherlands) at the discretion of the participant and the endoscopist. In case of poor bowel preparation the colonoscopy was interrupted and
re-scheduled. Withdrawal time was at least 6 minutes. All detected lesions were removed during the same procedure if possible. Pathological assessment of tissue samples provided a definitive diagnosis. Advanced neoplasia was defined as at least one CRC or advanced adenoma, adenoma of 10 mm or larger, ≥ 25% villous histology or high-grade dysplasia.

Risk factors

Based on a review of the literature, a set of putative risk factors for FIT positivity and negativity was selected. This set included only variables that could be collected through medical history and questionnaires, without any additional testing and collected in all patients. The definitive set consisted of data on age, sex, CRC family history, current smoking, history of smoking, BMI, regular Aspirin or NSAID use, total calcium intake and physical activity. Smoking was defined as smoking of cigarettes or cigars at the time of answering the questionnaire. These variables were considered potential risk factors for a false negative or a false positive FIT result. For risk factors for false positive results we used the same set of variables, completed by the presence of hemorrhoids and the use of aspirin.

Risk factor information was collected with a ten item questionnaire, which was based on three existing validated questionnaires: the Prevention Compass, the Municipal Health Agency, and the Interheart questionnaire. The questionnaire was handed out to the participants after arrival in the hospital and completed and collected immediately before the screening colonoscopy.

Statistical analysis

The FIT positivity threshold was set at 50 ng/ml. A false negative was defined as an individual with a negative FIT result in whom advanced neoplasia were detected during colonoscopy. A false positive test was defined as an individual with a positive FIT result and no advanced adenoma detected during colonoscopy.

We calculated the risk of a false negative result using logistic regression, with the variables in the risk factor questionnaire mentioned earlier as predictors. Risk factors were expressed as odds ratios, with 95% confidence intervals. Odds ratios for continuous variables were estimated using univariable logistic regression analyses, assuming the relation to be linear. A similar analysis was done for a false positive FIT result.

Missing data in the questionnaires were handled by multiple imputation. In multiple imputation, missing values are estimated from other related variables in the dataset. Several complete datasets are created, in which different imputations are based on a random draw from different estimated underlying distributions.

To put the risk factors into perspective, we evaluated whether there were any...
significant differences in FIT sensitivity and FIT specificity between subgroups, as defined by the variables in the risk factor questionnaire. For this purpose we estimate risk factors for a false negative FIT result in the subgroup of participants with advanced neoplasia only (for differences in sensitivity) and risk factors for a false positive FIT result in the subgroup of participants without advanced neoplasia only (for differences in specificity).

In our analyses, we focused on statistically significant differences, using a significance level of 0.05, as well as on variables for which the estimated odds ratio exceeded 1.25. This second criterion was added since the group with false positives was anticipated to be larger than the group of false negatives, and because it is known that the number of screening participants with advanced neoplasia would be much smaller than the group without. All authors had access to the study data and had reviewed and approved the final manuscript. We used the STARD statement to check the completeness of our study report.11

Results
Colonoscopy results
The results of colonoscopy and of FIT have been presented before.8,10 In brief, of the 1,426 screening participants in the colonoscopy arm, 1,112 (78%) completed both the FIT and the questionnaire (Figure 1). Table 1 summarizes the background characteristics of the study group. Their mean age of participants was 61 and 52% was male. At colonoscopy, 101 participants (8.9%) had advanced neoplasia. Of these, 7 (0.6%) participants had CRC and 94 (8.3%) had at least one advanced adenoma. Overall, 102 participants had a positive FIT result (cut of 50ng/ml), 65 (65.7%) participants had a false negative FIT result and 66 (65.7%) had a false positive result.
Figure 1. Patient flow.

Eligible patients: n=1426

Excluded patients: No questionnaire and FIT test n=314

FIT n=1112

Abnormal result n=102

Colonoscopy n=102

Advanced neoplasia n=36

No advanced neoplasia n=66

Normal result n=1010

Colonoscopy n=1010

Advanced neoplasia n=65

No advanced neoplasia n=945
Table 1 Descriptive statistics of the study group and odds ratios for false negatives and false positives

<table>
<thead>
<tr>
<th>Risk Factor</th>
<th>Total screening population OR (95% CI)</th>
<th>Advanced Neoplasia OR (95% CI)</th>
<th>False Positive</th>
<th>Total screening population OR (95% CI)</th>
<th>No Advanced Neoplasia OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number</td>
<td>1112</td>
<td>65</td>
<td>66</td>
<td>1112</td>
<td>65</td>
</tr>
<tr>
<td>Sex</td>
<td>Male: 569 (51%)</td>
<td>34</td>
<td>1.05 (0.64-1.73)</td>
<td>29</td>
<td>1.25 (0.74-2.03)</td>
</tr>
<tr>
<td></td>
<td>Female: 543 (49%)</td>
<td>31</td>
<td>1.41 (0.61-3.25)</td>
<td>37</td>
<td>1.03 (0.98-1.07)</td>
</tr>
<tr>
<td>Age (per additional year)</td>
<td>&lt;60 years: 559 (50%)</td>
<td>24</td>
<td>1.04 (1.01-1.09)</td>
<td>30</td>
<td>1.03 (0.98-1.07)</td>
</tr>
<tr>
<td></td>
<td>&gt;60 years: 553 (50%)</td>
<td>41</td>
<td>0.99 (0.92-1.06)</td>
<td>36</td>
<td>0.99 (0.93-1.06)</td>
</tr>
<tr>
<td>BMI kg/m²</td>
<td>&lt;20 kg/m²: 26 (2%)</td>
<td>1</td>
<td>0.99 (0.93-1.06)</td>
<td>1</td>
<td>0.99 (0.93-1.06)</td>
</tr>
<tr>
<td></td>
<td>20-25 kg/m²: 348 (31%)</td>
<td>22</td>
<td>0.96 (0.86-1.07)</td>
<td>28</td>
<td>0.97 (0.92-1.03)</td>
</tr>
<tr>
<td></td>
<td>25-30 kg/m²: 478 (43%)</td>
<td>33</td>
<td>0.99 (0.93-1.06)</td>
<td>24</td>
<td>0.97 (0.92-1.03)</td>
</tr>
<tr>
<td></td>
<td>&gt;30 kg/m²: 174 (16%)</td>
<td>9</td>
<td>0.96 (0.86-1.07)</td>
<td>11</td>
<td>0.97 (0.92-1.03)</td>
</tr>
<tr>
<td>Pack years smoking</td>
<td>&lt;24 years: 84 (8%)</td>
<td>6</td>
<td>1.01 (0.98-1.04)</td>
<td>5</td>
<td>1.00 (0.97-1.03)</td>
</tr>
<tr>
<td></td>
<td>&gt;24 years: 66 (6%)</td>
<td>6</td>
<td>1.07 (0.99-1.16)</td>
<td>8</td>
<td>1.00 (0.97-1.03)</td>
</tr>
<tr>
<td>Smoking status</td>
<td>Current smoker: 162 (15%)</td>
<td>16</td>
<td>2.02 (1.12-3.64)</td>
<td>52</td>
<td>1.63 (0.88-3.02)</td>
</tr>
<tr>
<td></td>
<td>Former or never smoker: 950 (85%)</td>
<td>49</td>
<td>1.03 (0.39-2.75)</td>
<td>14</td>
<td>1.00 (0.97-1.03)</td>
</tr>
</tbody>
</table>
### Table 1: Descriptive statistics of the study group and odds ratios for false negatives and false positives

<table>
<thead>
<tr>
<th>Risk Factor</th>
<th>Total screening population</th>
<th>OR (95% CI)</th>
<th>OR (95% CI)</th>
<th>OR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Advanced Neoplasia</td>
<td></td>
<td>1.05 (0.64-1.73)</td>
<td>1.41 (0.61-3.25)</td>
<td>1.25 (0.74-2.03)</td>
</tr>
<tr>
<td>Number</td>
<td></td>
<td>11126</td>
<td>56</td>
<td>6</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td>Male</td>
<td>Female</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>569 (51%)</td>
<td>543 (49%)</td>
<td></td>
</tr>
<tr>
<td>Age (per additional year)</td>
<td></td>
<td>&lt;60 years</td>
<td>&gt;60 years</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>559 (50%)</td>
<td>553 (50%)</td>
<td></td>
</tr>
<tr>
<td>BMI kg/m²</td>
<td></td>
<td>&lt;20 kg/m²</td>
<td>20-25 kg/m²</td>
<td>25-30 kg/m²</td>
</tr>
<tr>
<td></td>
<td></td>
<td>26 (2%)</td>
<td>348 (31%)</td>
<td>478 (43%)</td>
</tr>
<tr>
<td>Pack years smoking</td>
<td></td>
<td>&lt;24 years</td>
<td>&gt;24 years</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>84 (8%)</td>
<td>66 (6%)</td>
<td></td>
</tr>
<tr>
<td>Smoking status</td>
<td></td>
<td>Current</td>
<td>Former or never</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>162 (15%)</td>
<td>950 (85%)</td>
<td></td>
</tr>
<tr>
<td>Fiber intake (gram)</td>
<td></td>
<td>&lt;41 gram</td>
<td>&gt;41 gram</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>651 (59%)</td>
<td>461 (42%)</td>
<td></td>
</tr>
<tr>
<td>Calcium intake (mg)</td>
<td></td>
<td>&lt;1200 mg</td>
<td>&gt;1200 mg</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>985 (89%)</td>
<td>127 (11%)</td>
<td></td>
</tr>
<tr>
<td>NSAID</td>
<td></td>
<td>Regular User</td>
<td>Non-user</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>235 (21%)</td>
<td>877 (79%)</td>
<td></td>
</tr>
<tr>
<td>Aspirin</td>
<td></td>
<td>Regular user</td>
<td>Non regular user</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>175 (16%)</td>
<td>937 (84%)</td>
<td></td>
</tr>
<tr>
<td>Hemorrhoids</td>
<td></td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>63 (6%)</td>
<td>1037 (93%)</td>
<td></td>
</tr>
<tr>
<td>Diverticulosis</td>
<td></td>
<td>Yes</td>
<td>No</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>15</td>
<td>51</td>
<td></td>
</tr>
</tbody>
</table>
False Negative FIT result
Older participants (OR 1.04 per additional year) and smokers (OR 2.02) had a significantly higher risk of a false negative FIT result (see Table 1). Calcium intake had a small protective effect (OR 0.99). The other risk factors were not significantly associated with a false negative FIT, all other estimated odds ratios were smaller than our criterion of 1.25.

In our analysis of sensitivity modifiers, evaluated in the subgroup of patients with advanced neoplasia, we did not find significant associations with a false negative FIT result for any of the evaluated risk factors. For one factor the effect was higher than our preset criterion of 1.25: females with advanced neoplasia had a higher risk of a false negative FIT result, hence a lower sensitivity (OR 1.41).

False Positive FIT result
None of the risk factors evaluated were significantly associated with false positivity. For three variables, the estimated odds ratios were above our threshold: males were at increased risk of a false positive (OR 1.25), so were smokers (OR 1.63) and regular NSAID users (OR 1.32).

In our analysis of specificity modifiers, evaluated in the subgroup of patients without advanced neoplasia, we did not find significant associations between a false positive FIT result for any of the risk factors. When using our predefined threshold, specificity was found to be lower, though not significantly, in males (OR for a false positive: 1.25), smokers (OR 1.77) and in regular NSAID users (OR 1.30).

Since we observed that smokers were more likely to have a false negative and more likely to have a false positive result we compared the area under the ROC curve (AUC) in smokers and non-smokers (Figure 2). The AUC was lower in smokers (0.662 vs 0.706), though not significantly so (p=0.54).
Discussion

In this study on risk factors for false negative and false positive FIT results in screening we observed that the chances of having a false negative FIT were higher in smokers and in people at advanced age, both known risk factors for advanced neoplasia. Sensitivity for FIT was lower for women. For FIT false positivity no significant associations were observed, although this risk was increased in men, regular NSAID users and in smokers; in these groups specificity was also lower.

Strengths of this study are that all participants underwent colonoscopy examination, that the study included a random sample from the Dutch population, and the high response rate for the questionnaires, with almost ninety percent of participants completing the risk factor questionnaire. In addition, the distribution of life style factors in our study group is representative of that in the general Dutch population.\textsuperscript{29,30}
A number of other methodological issues need to be addressed. The sample size of this study was set at about 1,200 participants; there were 65 individuals with a false negative test result and 66 others had a false positive test result. These numbers limit the power to detect risk factors with a more modest association, especially for those with a low prevalence. For this reason we also considered risk factors with an odds ratio exceeding 1.25 worth mentioning. Although colonoscopy is the reference standard for colonoscopy in particular smaller lesions may have been missed, which would attenuate the associations observed. Furthermore, our study was conducted with self-administered questionnaires, a method that may be susceptible to socially desirable answers, though all participants were informed that the questionnaires were handled anonymously.

To our knowledge there is no literature available on risk factors associated with having a false negative FIT. For false positivity limited literature is available. Chiang et al. conducted a prospective cohort study, in which asymptomatic people age 18 years and older underwent a FIT test. They identified reasons for false positivity and found an association between false positivity and antiplatelet drugs. Morikawa et al. investigated FIT sensitivity in detecting small adenomas. They found that the false positive rate increased with age in men. Precise reasons were unknown, but they hypothesized that it might have been due to patients’ habits, like the use of alcohol use or antiplatelet drugs.

Our study shows that older individuals and smokers have a higher chance of a false negative test result. Age and smoking are both known risk factors for CRC. However, FIT sensitivity was not significantly lower among the older individuals and smokers.

The risk of having a false positive FIT result was higher in males, smokers and regular NSAID users. Here the overall effects were paralleled by lower specificities, which suggest that these effects on false positivity are differential. None of these effects were significant, however, so we offer these as hypotheses to be tested in further studies.

The findings from this study, when confirmed, could be used in targeting screening more towards groups that have a high risk for having a false negative test result. Especially in smokers this can have substantial implications. Individuals in this group have a higher chance of a false positive or a false negative test, and FIT sensitivity and specificity for FIT are lower, though the difference in accuracy was not significant. This, together with their higher chance of having CRC should be considered in screening projects. In breast cancer screening programs such differentiations have already been developed. Women with dense breasts are known to have an increased risk of having breast cancer. Breast density is a measurement of the ratio between radio sense epithelium and stroma to radiolucent fatty tissue, this radiologic finding is not related to the perceived density of breast tissue on palpation. With higher breast density, sensitivity is lower in mammographic screening. A risk model including breast density to identify women at increased risk for
breast cancer was developed. This might also be helpful for CRC screening. Like smoking and age, breast density is a risk factor for developing breast cancer as well as a risk factor for having a false negative screening test. This would justify the development of different screening regimes for these specific populations.

It is known that re-attendance in screening rounds is lower in individuals with a previous false positive screening test. In CRC screening a decision analysis has been presented on follow up of individuals with a false positive FIT. Haug et al concluded that repeated FIT screening 10 years after a false positive test is the optimal screening interval in terms of clinical benefit and required resources.

In conclusion, our study shows that the chances of having a false negative FIT are higher in smokers and in people at advanced age, both known risk factors for advanced neoplasia. Males, smokers and regular NSAID users have a higher chance of having a false positive test result.

FIT sensitivity was not significantly affected in smokers and in people at advanced age, subgroups with increased risk for advanced CR neoplasia but FIT sensitivity was lower in women. FIT specificity was lower in males, smokers and regular NSAID users. Our results can be used for further evidence based individualization of screening strategies.
References


