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The impact of immunochemical faecal occult blood testing on colorectal cancer incidence

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ABSTRACT

Background: The efficacy of colorectal cancer screening based on faecal immunochemical test, in terms of reduction of colorectal cancer incidence, is under debate. In the district of Florence, an organized screening programme based on faecal immunochemical test has been running since the early 1990s. The aim of this study was to compare the risk of developing colorectal cancer for subjects undergoing faecal immunochemical test with those who did not undergo the test in the same period.
Methods: Two cohorts were analyzed: subjects who underwent an initial faecal immunochemical test between 1993 and 1999 ("attenders"), and unscreened residents in the same municipalities invited to perform the faecal immunochemical test in the same period ("non-attenders"). Kaplan–Meier and Cox regression analysis were performed to evaluate the risk of developing colorectal cancer.
Results: The attenders' and non-attenders' cohorts included 6961 and 26,285 subjects, respectively. Cox analysis showed a reduction in colorectal cancer incidence of 22% in the attenders' compared to the non-attenders' cohort (hazard ratio = 0.78, 95% Confidence Interval: 0.65–0.93).
Conclusion: Our results support the hypothesis that screening based on a single faecal immunochemical test every 2 years produces a significant decrease in colorectal cancer incidence after an average follow-up observation period of 11 years.

1. Introduction

The effectiveness of colorectal cancer (CRC) screening programmes based on faecal immunochemical test (FIT) in reducing the incidence of CRC is an open issue. In the American Cancer Society guidelines for the early detection of cancer, FIT is classified as a test that primarily finds cancer [1]. So far, only the Minnesota Trial, which used a faecal occult blood test based on guaiac (gFOBT) [2], has shown a decrease in cancer incidence; however, the results were questioned due to the large proportion of people (around 40%) who underwent at least one colonoscopic examination as a consequence of a gFOBT-positive result. FIT is much more effective than gFOBT in detecting advanced adenomas [3]. Two randomized trials have demonstrated the efficacy of flexible sigmoidoscopy [4,5] in reducing CRC incidence by means of removing advanced adenomas. More recently, modelling-based approaches have predicted not only a significant reduction in cancer incidence for people undergoing FIT examinations [6], but also that 3 to 5 rounds of FIT (depending on participation) can detect the same number of subjects with advanced adenomas as a single round of sigmoidoscopy [7].

In the district of Florence, Italy, an organized CRC screening programme has been running since 1982. gFOBT was replaced by FIT at different times in each municipality over the course of 6 years. The reason for such a change derived from some studies demonstrating that FIT test increased sensitivity [8] and provided a better trade-off between sensitivity and specificity than gFOBT [9]. These results have been confirmed some years later in randomized clinical trials [3,10].

The effectiveness of the Florentine programme in reducing mortality for CRC has been evaluated both by a case-control study [8] and by comparison of areas with different start up of CRC screening programmes [11,12]. Recently, the trends analysis of CRC incidence [13] for the entire district during the period 1985–2005 for people aged 50–70 years showed an increase until 1996, and a subsequent significant decrease. This trend is different from that recorded in the rest of Italy, where an increase of CRC rates is still evident [14].

The aim of the present study was to analyze the risk of developing CRC in the 11 years following an initial FIT (carried out in the

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mid-1990s) and compare it with subjects who, although invited, did not undergo the test during the same period of time.

2. Materials and methods

2.1. Study population and screening programme

We considered the period from January 1, 1993 to December 31, 1999, in which FIT (single sample, biennial, age range 50–70 years) was progressively introduced in 24 Florentine municipalities. FIT testing was based on the reversed passive haemagglutination (RPHA) test (Immudia Hem SP, FujiRebio, Tokyo, Japan), developed at the 1/8 dilution according to the recommendations of the manufacturer. In 1996, new technologies provided by the manufacturer introduced a partial automation of the procedure concerning the dispensing and the reading phases. In 2000, based on the results of a comparative study, the RPHA was replaced by a latex agglutination test (OC-Hemodia, Eiken, Tokyo, Japan): a quantitative, completely automated FIT used on one-day sampling, positivity cut-off being set at 100 ng/ml of sample solution.

Over this period, the compliance to invitation was about 40% [15]. According to the screening protocol, all subjects were invited to the subsequent screening after 2 years, whether or not they had participated in the first round.

2.2. Definition of attenders and non-attenders

We defined as "attenders" the cohort of subjects, aged 50–70 years, residing in the study area, who had done their first FIT during the period 1993–1999 as a consequence of a screening invitation. The cohort of "non-attenders" included those who had been invited during the same period but who did not comply with the first invitation. Since the screening activity with FIT was progressively introduced during the enrolment phase (1993–1999), some municipalities (14) carried out more than one screening round in that period. In these cases, a person that did not comply with the first invitation could comply with the subsequent one. Due to the period of time elapsed from the first invitation until the screening test, we attributed these subjects to the non-attenders' cohort, in order to correctly attribute the exposure time to the non-attenders' cohort.

2.3. Statistical analysis

We linked the resident population of each municipality, at the time that FIT was introduced, with the screening database in order to define the cohorts of "attenders" and "non-attenders". We excluded all subjects who had undergone a gFOBT in the previous years, and those for whom a CRC had been diagnosed before the enrolment period. The 2 cohorts were then linked with the files of the Tuscan Cancer Registry according to the International Classification of Disease for Oncology (ICDO) codes 153.0–153.9, 154.0–154.2, and 154.4–154.9, in order to identify CRCs occurring in both cohorts. The follow-up period lasted from the date of invitation to the date of CRC diagnosis, death, emigration from the area, lost to follow-up, or the set end of follow-up (December 31, 2008), whichever came first. The date and the causes of death were obtained from the Tuscany Mortality Registry; the date of migration and vital status were identified by linkage with municipalities' registries.

We defined as cancers detected by screening those that occurred within 1 year after a positive test.

The 2 cohorts were followed up until December 31, 2008. We performed a first descriptive analysis with the Kaplan–Meier estimate. Furthermore, a Cox regression analysis, adjusted for age classes ($\leq$50, 55–59, 60–64, and $\geq$65) and sex, was performed to evaluate the risk of being diagnosed with a CRC in both the attenders' and not-attenders' cohorts.

The Cox model is a proportional hazard regression model based on the assumption that failure rates are proportional, given that the hazard ratio does not depend on time. In our analysis, proportionality of the hazards was not guaranteed due to the time cumulative hazard of the 2 cohorts. In fact, the risk of being diagnosed with CRC was higher in the first follow-up period (about 6 years) in the attenders' cohort, due to the early diagnosis during the screening activity, and was lower in the non-attenders' cohort. After this period the trend was inverted, causing an intersection of the hazard curves at the sixth year of follow-up (catch-up time).

For this reason, we divided the analysis into 2 periods according to the catch-up time, i.e. one period for the first 6 years and a second for the remaining years of follow-up. Within each period the proportion of hazard was guaranteed.

We computed a standardized incidence rate for the entire study period adjusted for sex and age in order to obtain the number of expected new cases that would have occurred in the attenders' cohort if it had had the same occurrence of CRCs as the non-attenders' cohort. We applied the sex and age-specific cancer incidence rates of the non-attenders' cohort to the person-times of the attenders' cohort to compute the number of expected cancer cases.

We also evaluated the effect of the screening according to cancer site by dividing CRCs between proximal (ICDO codes: 153.0, 153.1, 153.4–153.6, 153.8) and distal sites (ICDO codes: 153.2, 153.3, 153.7, 154.0–154.2, and 154.4–154.8). CRCs were excluded from this analysis when it was impossible to identify the exact site of occurrence (not specified 153.9 and 154.9).

Finally, we compared the mortality rates from CRC between the 2 cohorts, in order to estimate the effect of FIT screening on CRC mortality. We computed the standardized mortality ratio (SMR) considering the European standard population as reference. The 95% confidence intervals for the SMR have been reported.

All statistical analyses have been performed using the STATA software version 12.1 [16].

3. Results

The study population (Fig. 1) consisted of 60,040 subjects aged 50–70 years and resident in the area under study. Three-hundred and thirty-six people who had been diagnosed with a CRC before their invitation to FIT were excluded. Overall 59,704 subjects were invited to the first FIT examination. Among them, 24,240 (40.6%) performed the FIT test and 35,464 (59.4%) did not comply to the invitation. From the present study we excluded 17,279 subjects from the attenders' cohort, and 9179 from the non-attenders' cohort who had performed a previous gFOBT. At the end, the attenders' and non-attenders' cohorts under study included 6961 and 26,285 subjects, respectively. Small differences between the 2 cohorts in term of sex and age distribution were present. Among the non-attenders there were more males and older subjects than among the attenders: in fact, there were 3542 women in the attenders' cohort (50.8%) and 13,137 in the non-attenders' cohort (49.9%) (Pearson chi-squared = 1.18, $p = 0.18$). The mean age at entry into the study was 58.3 years (standard error (SE) = 0.07) for the attenders' cohort and 58.8 years (SE = 0.04) for the non-attenders' cohort (t-test = -6.99, $p < 0.005$). From the date of FIT introduction until the end of follow-up, an average of 5 screening invitations at two-yearly intervals were offered to all enrolled subjects; screened subjects performed, on average, 3.6 FITs. About 21% of the subjects underwent only 1 test, 15% underwent 2 tests, and 64% more than 2 tests. After the year 1999, 10% of the subjects in the non-attenders' cohort performed only 1 test, and less than 10% performed 2 or more tests.

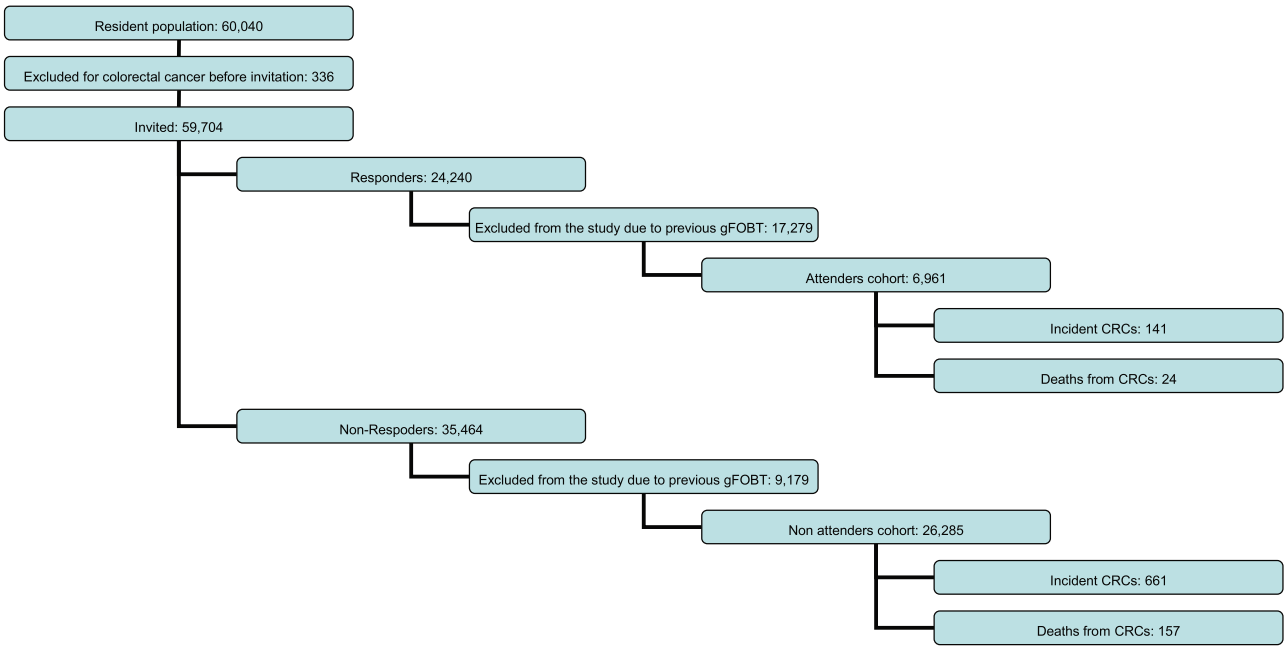

Fig. 1. Flow-chart describing the study design.

The mean positivity rate for the attenders' cohort was 5.4% at first tests and 3.5% at subsequent tests. On average, only 72.7% of positive subjects actually underwent colonoscopy. In the same period, 141 and 661 CRCs occurred in the attenders' and non-attenders' cohorts, respectively. Among the 141 CRCs occurred in the attenders' cohort, 44 (31.2%) resulted screening-detected. The mean follow-up period was 11.2 years (SD = 2.5) and 10.6 years (SD = 3.0) in the attenders' (78,024 person years) and non-attenders' (277,878 person years) cohorts, respectively. Ninety-seven (0.3%) subjects migrated from the study area, and 323 subjects (0.97%) were lost to followup. The Kaplan–Meier estimates (Fig. 2) showed an initial cumulative CRC risk of the attenders' cohort higher than that of the non-attenders' cohort; the CRC risk equalized at about 6 or 7 years, and thereafter became significantly lower.

The Cox model (Table 1), adjusted for sex and age, showed an overall statistically significant reduction in CRC incidence of 22% (HR = 0.78, 95%CI: 0.65–0.93) in the attenders' versus the non-attenders' cohort.

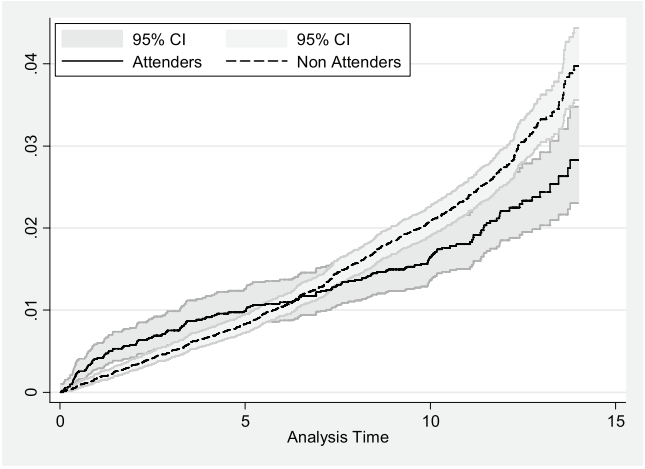

Fig. 2. Colorectal cancer Cumulative Incidence (%) for the attenders' (6961 subject and 78,027 person years) and non-attenders' (26,285 subject and 277,878 person years) cohorts.

Table 1
Hazard ratios of developing a Colorectal Cancers for attenders versus non-attenders subjects. Cox model during the entire time period.

	HR	95% CI
Non-attenders	Ref.	
Attenders	0.78	0.65–0.93
Males	Ref.	
Females	0.55	0.48–0.63
≤54 years	Ref.	
55–59 years	1.39	1.12–1.73
60–64 years	1.67	1.35–2.06
≥65 years	2.60	2.14–3.17

As expected, we observed an excess of risk in the attenders' cohort (not statistically significant) during the catch-up period (i.e. the first 6 years), and thereafter a significant reduction of CRC incidence in the attenders' cohort in comparison with the non-attenders' cohort ($HR_{\text{first 6 years}} = 1.06$, 95% CI: 0.82–1.37), ($HR_{\text{after the 6th year}} = 0.60$, 95% CI: 0.46–0.79).

The standardized incidence ratio analysis showed that, in contrast with the 141.0 CRCs observed among the 6961 attenders, we would have expected 180.2 CRC cases; thus, for every 1000 people screened, the screening activity had prevented about 5.6 CRCs.

No statistically significant difference was observed in terms of a possible protective effect of the cancer site (HR = 0.76; 95% CI: 0.6–0.9 for distal cancers, and HR = 0.79; 95% CI: 0.6–1.1 for proximal cancers). No differences were observed when taking into account the possible protective effect of sex or age at entry (data not shown).

Comparison of the mortality for CRCs in the 2 cohorts showed a significant reduction of CRC mortality in the attenders' cohort as compared to the non-attenders' cohort (SMR = 0.59, 95% CI: 0.37–0.93).

The Kaplan–Meier estimates (Fig. 3) showed the cumulative risk of death from CRC of the attenders' and non-attenders' cohorts. The mortality risk seemed to be similar in the 2 cohorts for the first 5 years of follow-up; thereafter the risk of the non-attenders increased more rapidly than that of the attenders.

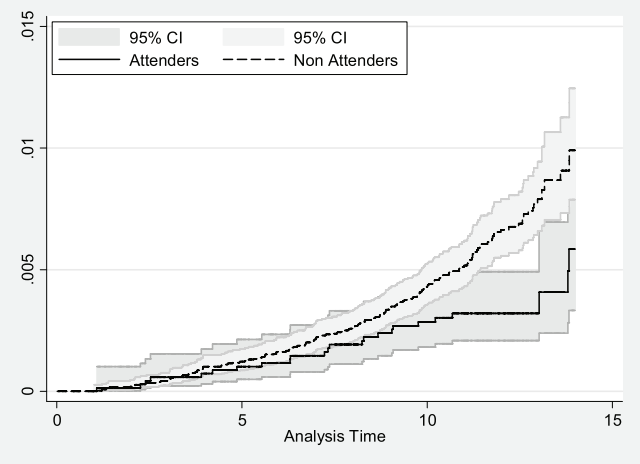

Fig. 3. Colorectal cancer Cumulative Mortality (%) for the attenders' (6961 subject and 80,808 person years) and non-attenders' (26,285 subject and 286,675 person years) cohorts.

4. Discussion

As far as we are aware, this observational study is the first analysing the effect on CRC incidence of a FIT-based screening programme. We compared our 2 cohorts (attenders and non-attenders) on the basis of the compliance to a first screening round using FIT: an intervention cohort of subjects screened in the first round of FIT (when FIT was introduced), and a comparison cohort of subjects resident in the same area and invited to the screening in the same period but who did not perform the screening test during the period 1993–1999.

Regarding the comparability of the 2 cohorts, they differed for age and sex, but these 2 confounders were adjusted in our analysis. The study cohorts were very similar with regard to previous examinations. In fact, we excluded from the analysis all the subjects who had performed a guaiac test before the study period. We do not know whether the baseline risk for developing a CRC is similar for attenders and non-attenders. In UK, individuals who decline an invitation to undertake FOBt screening exhibit a range of lifestyle factors that can potentially increase their risk of CRC [17].

In Italy some studies reported inconsistent results about the socio-economic and educational status related to CRC risk. Faggiano et al. [18] reported a low risk of colon cancer associated with low social strata, whereas Spadea et al. [19] reported an increased risk of CRC for subjects with a low educational level. Finally, the most recent publication of the PASSI study [20], reported no differences in CRC screening participation rates associated with different socio-economic status.

In a previous study [12], carried out in the same area and period as ours, the risk factors for CRC (age, place of birth, educational level, body mass index, food and wine consumption, smoking habits) were not statistically different in the compliers and not compliers, except for the factor "familial risk", which was higher among complying subjects. In the same study, compliers were slightly more likely to report other diagnostic intestinal examinations performed outside the programme than non-compliers (12.4% for compliers versus 9.2% for non-compliers). However, this difference was small (3.2%) and far from statistical significance ($P = 0.18$).

Finally, also the analysis of CRC mortality seems to confirm the comparability of the 2 cohorts in terms of basic risk for CRC. Overall, we observed a reduction in CRC mortality of 41% for the attenders as compared to the non-attenders. This reduction is in accordance with 2 case-control studies [8,21] where, as in the present analysis, attenders were compared to non-attenders. What is worth noting is the pattern of the cumulative trend. In fact, if the non-attenders' cohort had experienced a higher risk of CRC, a higher mortality should be expected from the beginning, whereas no differences were observed in the first 5 years of the study. In the following years the decrease in CRC mortality can be attributed mainly to the CRC early diagnosis and, partially, in the last period, to a decreased incidence in the attenders' cohort.

It is therefore not likely that the attenders were at lower risk for CRC. Concerns regarding a possible healthy-volunteer effect could be raised. Such an effect would exist in our case if there was a relationship between compliance to screening and lower prevalence of CRC cancer. Two different situations have to be considered. First, some individuals did not attend the FIT as they were on a diagnostic path of assessment for CRC at the time of invitation. If this were the case we would overestimate the protective effect of screening on CRC incidence. In order to estimate the potential effect of such a bias, we excluded all cancers that occurred in the first 6 months after invitation only in the control group. With this condition, the relative protective effect decreased slightly from 0.78 (95% CI: 0.65–0.93) to 0.80 (95% CI: 0.67–0.96), but maintained its statistical significance. Moreover, we observed the same pattern of a 40% CRC incidence reduction after 6–7 years. Second, it could also be possible that some subjects attended the screening due to the presence of symptoms for CRC. In this case, we would have underestimated the protective effect of the screening on CRC incidence. As a matter of fact, 31% of the individuals positive to FIT reported previous bleeding in the history collected before colonoscopy. Additionally, it is worth noting that in those subjects the Positive Predictive Value (PPV) for cancer was almost double than that of subjects not reporting a previous bleeding (PPV 17.4%; 95% CI: 10.1%–27.1% for subjects reporting bleeding, and 9.7%; 95%CI: 5.8%–14.8% for subjects not reporting bleeding; p value = 0.07).

In conclusion, a healthy-volunteer effect did not play a major role on the observed decrease of CRC incidence.

Another important point in our discussion concerns the fact that in the subsequent screening rounds (i.e. after 1999) 24% of non-attenders actually underwent a CRC screening test (average number of tests = 0.5). In the overall evaluation this slight contamination of the comparison cohort should also be considered, although it had little impact since the tests were performed after 1999. These 2 arguments (namely supposed familial risk in screened subjects and a slight contamination of the non-attenders cohort) indicate that the observed difference probably underestimated in some way the real effect of FIT-based screening on CRC incidence.

Our results can be compared with those emerging from 2 randomized trials of "once only flexible sigmoidoscopy" (the UK multicentre trial, by Atkin et al., [4] and the Italian SCORE trial, by Segnan et al. [5]), which reported a significant reduction of CRC occurrence in their active groups. A correct comparison should be made considering the "per-protocol" analysis of those trials (i.e., the comparison of attending versus not-attending subjects). In the flexible sigmoidoscopy trials, the reduction in CRC occurrence was greater than that observed in the present study (33% in the UK study and 31% in the SCORE trial). It is worth mentioning that the shapes of the incidence rate curves in the flexible sigmoidoscopy trials and our study are similar. In fact, while we initially observed an increase in CRC incidence in the first years, due to the early diagnosis of prevalent CRCs, after a period of about 4–5 years in the flexible sigmoidoscopy trials and of 6–7 years in the present study, we observed a dramatic decrease in CRC incidence for the attenders. A further reduction of CRC incidence in the attenders' cohort, either in absolute or relative terms, would probably be observed with a longer follow-up.

Moreover, the fact that the protective effect of FIT is similar for both left and right-sided CRC makes the use of FIT potentially

interesting as complementary test in the years after a negative sigmoidoscopy.

In conclusion, our results support the hypothesis that the implementation of CRC screening based on a single FIT every 2 years, with a cut-off of 100 ng/ml, reduces CRC incidence even with only a few screening rounds. In fact, this result was obtained despite a low compliance to colonoscopy (73%). Thus the benefit could also have been more important with a higher compliance to colonoscopy.

Recently, several studies [22–24] evaluated the impact of using different FIT positivity cut-offs and single vs multiple sampling in terms of sensitivity/specificity ratio. In particular, the Italian study [23] suggested that lowering the FIT cut-off influences the detection rate of advanced adenomas more than the detection of cancers. Therefore, it is likely that the use of a modern quantitative immunochemical test with a low positivity cut-off or multiple samples may produce an even greater reduction of CRC incidence.

Conflict of interest statement

All authors declare to have not conflict of interest.

References

[1] http://www.cancer.org/Healthy/FindCancerEarly/CancerScreeningGuidelines/american-cancer-society-guidelines-for-the-early-detection-of-cancer

[2] Mandel JS, Church TR, Bond JH, et al. The effect of faecal occult-blood screening on the incidence of colorectal cancer. The New England Journal of Medicine 2000;343:1603–7.

[3] van Rossum LG, van Rijn AF, Laheij RJ, et al. Random comparison of guaiac and immunochemical faecal occult blood tests for colorectal cancer in a screening population. Gastroenterology 2008;135:82–90.

[4] Atkin WS, Edwards R, Kralj-Hans I, et al., UK Flexible Sigmoidoscopy Trial Investigators. Once-only flexible sigmoidoscopy screening in prevention of colorectal cancer: a multicentre randomized controlled trial. Lancet 2010;375:1624–33.

[5] Segnan N, Armaroli P, Bonelli L, et al., SCORE Working Group. Once-only sigmoidoscopy in colorectal cancer screening: follow-up findings of the Italian randomized controlled trial–SCORE. Journal of the National Cancer Institute 2011;103:1310–22.

[6] Lansdorp-Vogelaar I, van Ballegooijen M, Zauber AG, et al. Effect of rising chemotherapy costs on the cost savings of colorectal cancer screening. Journal of the National Cancer Institute 2009;101:1412–22.

[7] Ventura L, Zappa M, Carreras G, et al. What is the best screening strategy to detect advanced colorectal adenomas? Simulation from ongoing Italian screening experiences. Tumori 2011;97:547–50.

[8] Zappa M, Castiglione G, Grazzini G, et al. Effect of faecal occult blood testing on colorectal mortality: results of a population-based case-control study in the district of Florence, Italy. International Journal of Cancer 1997;73:208–10.

[9] Castiglione G, Zappa M, Grazzini G, et al. Immunochemical vs guaiac faecal occult blood tests in a population-based screening program for colorectal cancer. British Journal of Cancer 1996;74:141–4.

[10] Hol L, van Leerdam ME, van Ballegooijen M, et al. Screening for colorectal cancer: randomised trial comparing guaiac-based and immunochemical faecal occult blood testing and flexible sigmoidoscopy. Gut 2010;59:62–8.

[11] Costantini AS, Martini A, Puliti D, et al. Colorectal cancer mortality in two areas of Tuscany with different screening exposures. Journal of the National Cancer Institute 2008;100:1818–21.

[12] Zappa M, Castiglione G, Giorgi D, et al. Re: Participation in colorectal cancer screening: a review. Journal of the National Cancer Institute 1998;90:465.

[13] Crocetti E, Buzzoni C, Zappa M. Colorectal cancer incidence rates have decreased in central Italy. European Journal of Cancer Prevention 2010;19:424–5.

[14] http://itacan.ispo.toscana.it/italian/itacan.htm. (28.03.2013).

[15] Grazzini G, Castiglione G, Isu A, et al. Colorectal cancer screening by faecal occult blood testing: results of a population-based experience. Tumori 2000;86:384–8.

[16] StataCorp. Stata Statistical Software: Release 12. College Station, TX: StataCorp LP; 2011.

[17] The UK CRC Screening Pilot Evaluation Team. Evaluation of the UK Colorectal Cancer Screening Pilot. Final Report, 2003.

[18] Faggiano F, Partanen T, Kogevinas M, et al. Socioeconomic differences in cancer incidence and mortality. IARC Scientific Publications 1997;138:65–176.

[19] Spadea T, D'Errico A, Demaria M, et al. Educational inequalities in cancer incidence in Turin, Italy. European Journal of Cancer Prevention 2009;18:169–78.

[20] Rapporto nazionale Passi 2011: lo screening per il cancro del colon retto.

[21] Saito H, Soma Y, Koeda J, et al. Reduction in risk of mortality from colorectal cancer by faecal occult blood screening with immunochemical hemagglutination test. A case-control study. International Journal of Cancer 1995;16:61, 465-9.

[22] van Rossum LGM, van Rijn AF, Laheij RJF, et al. Cutoff value determines the performance of a semi-quantitative immunochemical faecal occult blood test in a colorectal cancer screening program. British Journal of Cancer 2009;101:1274–81.

[23] Grazzini G, Visioli CB, Zorzi M, et al. Immunochemical faecal occult blood test: number of samples and positivity cutoff. What is the best strategy for colorectal cancer screening? British Journal of Cancer 2009;100:259–65.

[24] Guittet L, Bouvier V, Mariotte N, et al. Comparison of a guaiac based and an immunochemical faecal occult blood test in screening for colorectal cancer in a general average risk population. Gut 2007;56:210–4.