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Microsatellite instability has a positive prognostic impact on stage II colorectal cancer after complete resection: results from a large, consecutive Norwegian series

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Background: Microsatellite instability (MSI) was suggested as a marker for good prognosis in colorectal cancer in 1993 and a systematic review from 2005 and a meta-analysis from 2010 support the initial observation. We here assess the prognostic impact and prevalence of MSI in different stages in a consecutive, population-based series from a single hospital in Oslo, Norway.

Patients and methods: Of 1274 patients, 952 underwent major resection of which 805 were included in analyses of MSI prevalence and 613 with complete resection in analyses of outcome. Formalin-fixed tumor tissue was used for PCR-based MSI analyses.

Results: The overall prevalence of MSI was 14%, highest in females (19%) and in proximal colon cancer (29%). Five-year relapse-free survival (5-year RFS) was 67% and 55% ($P = 0.030$) in patients with MSI and MSS tumors, respectively, with the hazard ratio (HR) equal to 1.60 ($P = 0.045$) in multivariate analysis. The improved outcome was confined to stage II patients who had 5-year RFS of 74% and 56% respectively ($P = 0.010$), HR = 2.02 ($P = 0.040$). Examination of 12 or more lymph nodes was significantly associated with proximal tumor location ($P < 0.001$).

Conclusions: MSI has an independent positive prognostic impact on stage II colorectal cancer patients after complete resection.

Key words: adenocarcinoma, colorectal neoplasms, lymph nodes, microsatellite instability, prevalence, prognosis

introduction

Colorectal cancer is among the most common malignancies in the western world [1] and is becoming more common in developing countries as they approach a western lifestyle [2]. In Norway, the age-adjusted incidence rate has doubled over the last 50 years and is now among the highest in Europe [3].

Several clinical and pathological factors have prognostic impact on colorectal cancer including tumor stage, residual tumor (R-) status [4], tumor differentiation [5, 6], bowel perforation and emergency surgery [7]. In colon cancer, the number of examined lymph nodes has a prognostic impact [8–11]. Risk stratification according to these clinicopathological factors is applied to select patients for (neo-) adjuvant treatment. In Norway, stage III colon cancer patients with age less than 76 years are offered adjuvant chemotherapy. Stage II patients do not receive such therapy, except those with bowel perforation or less than nine examined lymph nodes after a thorough examination of the resected tissue. In rectal cancer, preoperative radiochemotherapy is recommended if the distance from the tumor or a metastatic lymph node to the mesorectal fascia is ≤3 mm, evaluated by magnetic resonance imaging.

However, current risk stratification does not adequately identify patients with good and poor prognosis. The 5-year relative survival rate of stage III colon cancer patients was 57% before adjuvant chemotherapy became standard treatment [3], which implies that more than half of these patients are cured by surgery alone and are over-treated when given adjuvant therapy. Five-year relative survival in stage II colon cancer is 75% [12], indicating that 25% of the patients relapse and die of cancer within 5 years after surgery. Possibly, adjuvant therapy for high-risk stage II patients might improve these results. Several biomarkers have been proposed to improve the identification of patients at risk of relapse, but none are implemented in clinical practice [13].

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Approximately 15% of all colorectal cancers display microsatellite instability (MSI), a molecular phenotype caused by defect mismatch repair [14–17]. In Lynch syndrome (former HNPCC), MSI is due to germline mutation in one of the MMR genes, usually *MLH1* or *MSH2* [18–20]. In sporadic colorectal cancer, MSI is mainly caused by epigenetic silencing of *MLH1* [21–23] and is characterized by poor differentiation, tumor-infiltrating lymphocytes, location in the proximal colon and association with female gender and age [14, 16, 17, 24–28].

We initially reported MSI as a marker of good prognosis in 1993 [14]. Subsequent reports have shown conflicting results; however, a systematic review from 2005 concluded that patients with MSI tumors have better prognosis than those with MSS tumors [29] and a meta-analysis from 2010 confirmed this finding [30]. It is yet to be decided whether this is valid for all stages, and the results from different studies differ at this point [24, 25, 28]. The aim of the present study was to evaluate the prognostic impact of MSI adjusted for stage and other clinical variables in a large, consecutive series from a single hospital.

materials and methods

Oslo University Hospital, Aker has a defined catchment area of 270 000 inhabitants. All patients with colorectal cancer admitted to the hospital in the period 1993–2003 were registered and clinical data recorded in a database. Registration has been controlled against the Norwegian Cancer Registry.

Major resection was defined as removal of the tumor-bearing bowel segment with the lymphovascular pedicle and mesentery with regional lymph nodes. Total mesorectal excision was carried out in all patients with rectal cancer. Fifteen percent of the patients underwent emergency surgery, due to obstruction or perforation of the bowel.

TNM-staging followed the UICC/AJCC system, version 5, for all patients. Based on the radiological examinations, intraoperative findings and macroscopic and microscopic examination, the resection was classified as R0 (complete resection/no residual tumor), R1 (microscopic residual cancer at the resection margin) or R2 (macroscopic or radiological evidence of residual cancer, locally or distant). For colon cancer, the total number of examined lymph nodes was registered.

The patients were split into three subgroups according to tumor location: proximal colon including the cecum through the transverse colon; distal colon including the left flexure through the rectosigmoid flexure; rectum was defined as the bowel up to 15 cm above the anal verge.

Colon cancer patients with age less than 76 years and all rectal cancer patients who underwent curative surgery entered a 5-year follow-up program (supplementary Table S1, available at *Annals of Oncology* online). Patients who were not enrolled in systematic follow-up would be admitted to our hospital if developing symptoms of relapse, implying that most relapses would be identified and registered. Information about death was retrieved from the Norwegian Tax Administration.

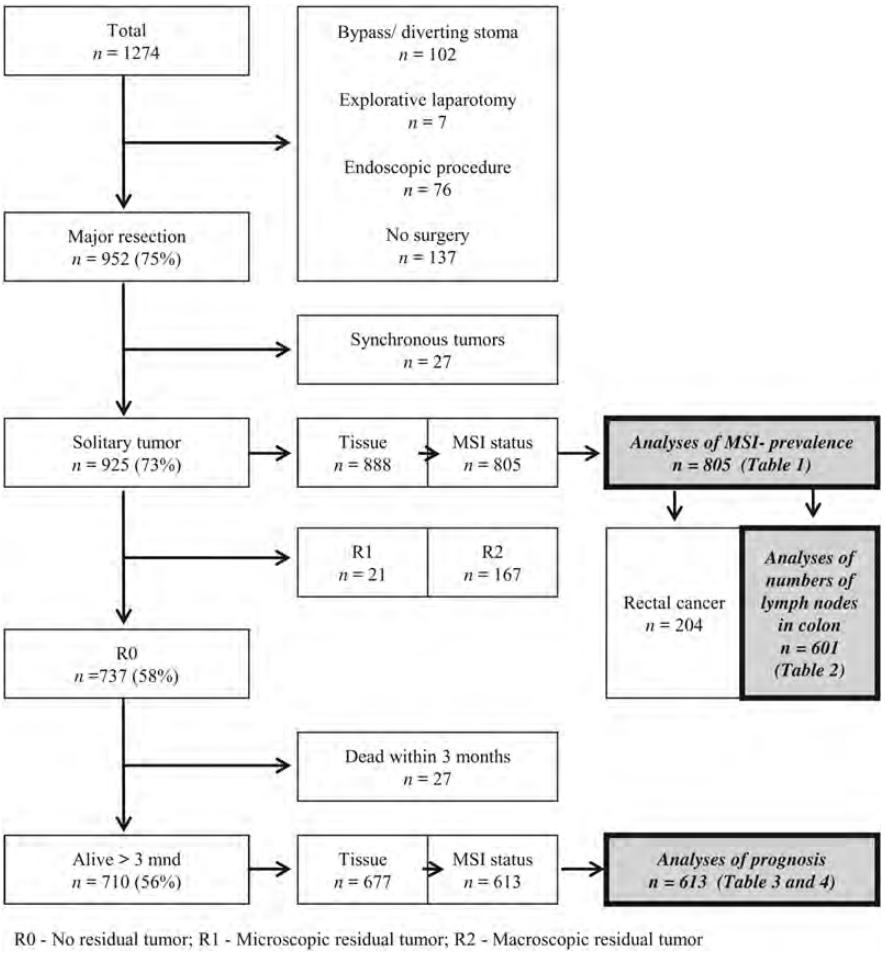


Figure 1. Flow chart for all patients with colorectal cancer admitted to Oslo University Hospital, Aker, in the period 1993–2003.

Formalin-fixed paraffin-embedded tumor tissue was retrieved for all patients who had undergone major resection, and HE sections were re-examined to confirm the presence of cancer and mark the most representative area. Four 25 μm sections were used for DNA extraction with QIAamp DNA Mini kit from Qiagen (GmbH, Hilden, Germany). The method was modified by adding an early step for removal of paraffin by heating to 90°C for 10 min after buffer was added.

For determination of the MSI status, microsatellite analyses were carried out for the five loci recommended by the National Cancer Institute [31]. PCR for the mononucleotides (BAT25 and BAT26) and the dinucleotides (D2S123, D5S346 and D17S250) were run separately. Both the reactions used 37 ng DNA templates in a 10 μl reaction volume consisting of a 1× Multiplex PCR Master mix (buffer, 1.5 mM $MgCl_2$, nucleotides and enzyme, QUIAGEN GmbH, Hilden, Germany), fluorescent primers and water. The mononucleotide markers underwent 30 cycles and the dinucleotide markers 35 cycles. Fragment analysis was accomplished on 3730 Genetic Analyzer (Applied Biosystems, Life Technologies, Carlsbad, California). Four DNA samples extracted from blood of healthy donors were included in each run as controls. The results were scored independently by two observers. The MSI status for each locus was determined after two independent runs with the same conclusion (MSI or wild type). If there were contradictory results, the locus was scored as 'not determined'. Samples with two or more loci exhibiting abnormal allelic ranges were scored as MSI high (MSI-H, from here on referred to as MSI). If one locus was MSI and four loci were wild type, the sample was scored as MSI low (MSI-L). Samples with wild type in all five loci were scored as microsatellite stable (MSS). For further analyses, MSI-L and MSS were included in the same group, and referred to as MSS, as were samples with four wild-type loci and one 'not determined' locus.

The associations between MSI, number of examined lymph nodes and different clinical variables were explored in contingency tables, and Pearson's chi-square test was applied. Logistic regression was used in multivariate models to explore different variables' impact on the MSI-status and the number of examined lymph nodes.

The prognostic impact of MSI and clinical variables was analyzed with 5-year overall survival (5-year OS) as primary endpoint; death from any cause was defined as event and patients were censored 5 years after surgery. The second endpoint was 5-year relapse-free survival (5-year RFS); deaths from any cause and recurrence (locally and/or distant) were defined as events [32]. The patients were censored at loss to follow-up, defined as the last date for clinical or radiological examination or at 5 years after surgery. Survival analyses were carried out using the Kaplan–Meier method, and the survival distributions were compared with the log-rank test. Multivariate analyses were carried out using Cox regression analyses, all

Table 1. Prevalence of MSI according to clinical and histopathological variables ($n = 805$)

Variables	Total	Univariate[a]		Multivariate[b]		
	N (%)	MSI *N* (%)	*P*	OR	95% CI	*P*
Total	805	112 (14)				
Sex						
Female	431 (54)	82 (19)	<0.001	Ref		
Male	374 (46)	30 (8)		0.41	0.24–0.70	0.001
Age						
<60 years	146 (18)	18 (12)	0.241	Ref		
60–70 years	164 (20)	16 (10)		0.42	0.18–1.00	0.051
70–80 years	300 (37)	46 (15)		0.61	0.30–1.24	0.174
>80 years	195 (24)	32 (16)		0.56	0.26–1.19	0.131
Tumor location						
Proximal colon	327 (41)	96 (29)	<0.001	Ref		
Distal colon	274 (34)	12 (4)		0.14	0.07–0.27	<0.001
Rectum	204 (25)	4 (2)		0.05	0.02–0.13	<0.001
Stage						
I	118 (15)	7 (6)	<0.001	Ref		
II	323 (40)	65 (20)		1.89	0.75–4.75	0.176
III	210 (26)	27 (13)		1.07	0.40–2.88	0.887
IV	154 (19)	13 (8)		0.83	0.17–4.03	0.818
Histopathologic grade						
G1 + G2	685 (85)	65 (10)	<0.001	Ref		
G3	102 (13)	42 (41)		7.34	4.06–13.27	<0.001
Mucinous	9 (1)	4 (44)		4.93	1.12–21.71	0.035
Surgery						
Elective	683 (85)	101 (15)	0.090	Ref		
Acute	122 (15)	11 (9)		0.44	0.21–0.95	0.038
Residual tumor						
R0	637 (79)	97 (15)	0.061	Ref		
R1	17 (2)	3 (18)		1.21	0.24–6.10	0.813
R2	151 (19)	12 (8)		0.37	0.10–1.46	0.157

[a]Contingency tables, chi-square test.

[b]Logistic regression, all included variables are displayed in the table.

variables from univariate analyses were entered into the models. A *P*-value of <0.05 was considered statistically significant. All analyses were carried out with SPSS 16.0 (IBM®SPSS®, IBM Corporation, Armonk, New York).

The study was carried out according to the Helsinki declaration and approved by the Regional Ethics Committee for Medical Research (REK approval 1.2005.1629) and the Norwegian Data Inspectorate.

results

The selection of patients included in the study is illustrated in Figure 1 and the characteristics of the cohorts included in the different analyses are displayed in the supplementary Table S2, available at *Annals of Oncology* online. A total of 1274 patients were admitted with colorectal cancer from 1993 to 2003 and 925 patients underwent major resection of a solitary tumor. Tumor tissue was available from 888 and the MSI status was successfully determined in 805 (91%) patients who were included in the analyses of MSI prevalence.

MSI prevalence and clinical variables

MSI was demonstrated in 112 (14%) patients (Table 1). MSI tumors were most frequent in the proximal colon and 86% of the MSI tumors were located proximal to the splenic flexure. MSI was more common in females who had a greater proportion of their tumors in the proximal colon (49% versus 31% in men, $P < 0.001$), but also had a higher frequency of MSI in their proximal tumors (34% versus. 20% in men, $P = 0.005$). The prevalence of MSI varied with tumor stage with the lowest frequency in stage I (6%) and the highest in stage II (20%). This was partly because stage I tumors were rare in the proximal colon ($n = 25$, 8%), whereas stage II tumors were frequent ($n = 145$, 44%). Including only proximal colon cancers, the frequencies of MSI in stage I ($n = 25$), stage II ($n = 145$), stage III ($n = 82$) and stage IV ($n = 75$) were 24%, 39%, 26% and 16%, respectively. MSI was most prevalent in tumors with poor differentiation (G3) and in mucinous tumors. In a multivariate analysis (Table 1), MSI was associated with female gender, tumor location in proximal colon, poor differentiation and elective surgery.

MSI and number of examined lymph nodes

In the analyses of number of lymph nodes, rectal cancer patients were excluded, leaving 601 colon cancer patients. Because of missing data for three patients, 598 patients were included in the analyses. Twelve or more examined lymph nodes were obtained in 31% of the patients and the

Table 2. Proportion of colon cancer patients with ≥12 examined lymph nodes (ln) according to clinical and histopathological variables ($n = 598$)

Variables	Total	Univariate[a]		Multivariate[b]		
		≥12 ln	*P*	OR	95% CI	*P*
	N (%)	*N* (%)				
Total	598	186 (31)				
MSI status						
MSI	108 (18)	46 (43)	0.004	Ref		
MSS	490 (82)	140 (29)		0.86	0.54–1.37	0.534
Sex						
Female	337 (56)	117 (35)	0.030	Ref		
Male	261 (44)	69 (27)		0.73	0.50–1.07	0.105
Age						
<60 years	92 (15)	41 (45)	0.019	Ref		
60–70 years	114 (19)	33 (29)		0.53	0.30–0.93	0.027
70–80 years	224 (38)	60 (27)		0.43	0.26–0.69	<0.001
>80 years	168 (28)	52 (31)		0.48	0.29–0.80	0.005
Tumor location						
Proximal colon	324 (54)	128 (40)	<0.001	Ref		
Distal colon	274 (46)	58 (21)		0.45	0.30–0.67	<0.001
Stage						
I	64 (11)	14 (22)	0.004	Ref		
II	249 (42)	78 (31)		1.60	0.95–2.68	0.075
III	153 (26)	63 (41)		2.50	1.44–4.35	0.001
IV	132 (22)	31 (24)		1.04	0.56–1.92	0.906
Histopathologic grade						
G1 + G2	498 (85)	155 (31)	0.903	Ref		
G3	83 (14)	27 (33)		0.87	0.50–1.50	0.611
Mucinous	8 (1)	3 (38)		1.11	0.25–4.86	0.888
Surgery						
Elective	482 (81)	153 (32)	0.284	Ref		
Acute	116 (19)	33 (28)		0.90	0.56–1.46	0.674

[a]Contingency tables, chi-square test.

[b]Logistic regression, all included variables are displayed in the table

distribution according to clinical variables is presented in Table 2. When including only tumors from the proximal colon ($n = 324$), the numbers of patients with 12 or more examined lymph nodes were 43 (45%) and 85 (37%) for MSI and MSS, respectively ($P = 0.203$). If only including MSS tumors ($n = 490$), the numbers with 12 or more lymph nodes were 85 (37%) and 55 (21%) for proximal and distal colon, respectively ($P < 0.001$). In multivariate analyses, age, tumor location and stage had a significant impact on the proportion with 12 or more examined lymph nodes, whereas the MSI status had no significant impact.

MSI and survival

The MSI status was successfully determined in 613 patients with solitary tumors who survived for >3 months after an R0-resection (Figure 1). These were included in the prognostic analyses, and matched well with all patients who underwent major resection with regard to age, gender and tumor location (supplementary Table S2, available at *Annals of Oncology* online). The group included 17 stage IV patients who underwent R0-resection of synchronous, distant metastases during or shortly after the primary operation.

Of the 613 patients included in the prognostic analyses, 157 (26%) experienced relapse and 224 (37%) died without known relapse. The 5-year estimated relapse rates were 10%, 23% and 42% in stages I–III, respectively according to the Kaplan–Meier method. For patients who survived without relapse, the median follow-up time was 65 months.

The 5-year OS rates were 69% and 61% for patients with MSI tumors and MSS tumors, respectively ($P = 0.214$), with the hazard ratio (HR) equal to 1.47 ($P = 0.112$). However, MSI was associated with significantly improved 5-year RFS (Table 3). Subgroup analyses demonstrated that the improved outcome for MSI tumors only applied to stage II, whereas no difference in the outcome was found in stage III (Figure 2). For stage I and IV, the numbers of MSI tumors were too small to draw any conclusions.

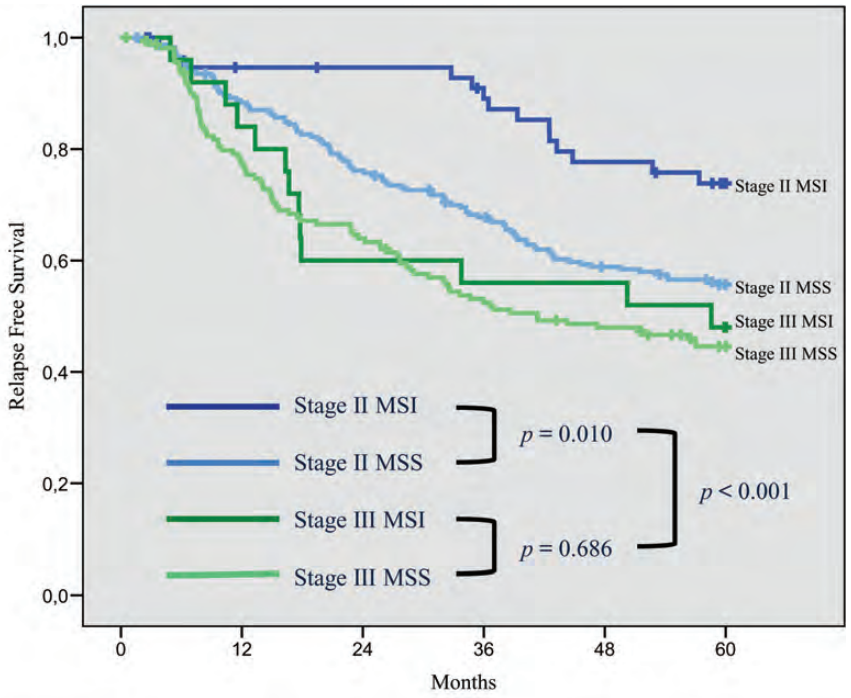
The prognostic impact of MSI status in stage II patients is presented in Table 4, showing 5-year RFS of 74% and 56% ($P = 0.01$) in MSI and MSS patients, respectively, with the HR equal to 2.02 ($P = 0.040$).

Table 3. Five-year relapse-free survival (5-year RFS) in stage I–IV colorectal cancer (R0-resection, solitary tumor, alive >3 months after surgery, $n = 613$)

Variables	Total N (%)	Univariate[a] 5-year RFS (%)	P	Multivariate[b] HR	95% CI	P
Total	613	56.5				
MSI status						
MSI	92 (15)	67.1	0.030	Ref		
MSS	521 (85)	54.7		1.60	1.01–2.52	0.045
Sex						
Female	321 (52)	58.3	0.488	Ref		
Male	292 (48)	54.6		1.10	0.85–1.43	0.451
Age						
<60	111 (18)	74.8	<0.001	Ref		
60–70	126 (21)	60.7		1.88	1.17–3.04	0.010
70–80	236 (39)	53.4		2.40	1.56–3.70	<0.001
>80	140 (23)	43.4		2.92	1.83–4.67	<0.001
Tumor location						
Proximal colon	238 (39)	59.5	0.284	Ref		
Distal colon	198 (32)	53.3		1.24	0.91–1.71	0.179
Rectum	177 (29)	56.1		1.51	1.07–2.13	0.019
Stage						
I	117 (19)	75.0	<0.001	Ref		
II	291 (48)	59.2		1.95	1.27–3.01	0.002
III	188 (31)	45.0		3.37	2.18–5.21	<0.001
IV	17 (3)	11.8		5.55	2.88–10.70	<0.001
Histopathologic grade						
G1/G2	534 (87)	58.2	0.025	Ref		
G3	66 (11)	45.1		1.84	1.24–2.73	0.003
Mucinous	7 (1)	57.1		1.31	0.42–4.15	0.642
Surgery						
Elective	544 (89)	58.2	0.004	Ref		
Acute	69 (11)	43.1		1.35	0.94–1.96	0.107

[a]Kaplan–Meier estimate, log-rank test.

[b]Cox Regression, all included variables are displayed in the table.



Time (months)		12	24	36	48	60
Stage II						
MSI (58)	Events	3	3	6	12	14
	At risk	52	51	47	41	35
MSS (233)	Events	27	55	74	94	101
	At risk	203	175	153	101	118
Stage III						
MSI (25)	Events	4	10	11	11	13
	At risk	21	15	14	14	11
MSS (163)	Events	36	57	75	82	87
	At risk	122	101	82	74	63

Figure 2. Five year relapse-free survival (RFS), stage II and III, $n = 479$.

discussion

The important finding in the present study was that stage II patients with MSI tumors have better outcome than patients with MSS tumors. This is in accordance with several other publications [24, 28–30, 33–35]. This was demonstrated in a large, consecutive and population-based series with minimal risk of selection bias. The comprehensive set of clinical data made it possible to adjust for several well-known prognostic factors. Patients with synchronous tumors were excluded because of the uncertainty regarding which tumor was most relevant for prognosis. We chose robust endpoints according to Punt et al. [32] and end points based on the cause of death were not considered due to the risk of bias due to erroneous cause of death. Analyses were restricted to 5-year survival, as most deaths after this time will not be cancer related. Patients were censored at the time of the last examination with regard to recurrence, and bias due to loss of follow-up was minimized. This report follows the recommendations for tumor marker prognostic studies [36]. Based on these conditions, the conclusion with regard to the prognostic impact of MSI is reliable.

The positive prognostic impact of MSI was confined to stage II patients. In contrast, Samowitz et al. found significant impact only in stage III patients in a study of 1000 colon cancer patients from California and Utah, all less than 79 years of age, and with different ethnic background [28]. Benatti et al. presented a series of 1263 colorectal cancer patients and found a positive prognostic impact of MSI in stage II and III [24]. Patients with clinical suspicion of hereditary colorectal cancer syndromes were also included in this study and the mean age was only 65 years. The prevalence of MSI was unusually high (20%). The current series has the advantage of not being biased by any selection among the enrolled patients.

From 1997, patients up to 75 years with stage III colon cancer receive 5FU-based adjuvant treatment. A systematic review with meta-analysis from 2009 reported that MSI tumors do not respond to this treatment [37] and this could

Table 4. Five-year relapse-free survival (5-year RFS) in stage II colorectal cancer (R0-resection, solitary tumor, alive > 3 months after surgery, $n = 291$)

Variables	Total N (%)	Univariate[a] 5-year RFS (%)	P	Multivariate[b] HR	95% CI	P
Total	291	59.2				
MSI status						
MSI	58 (20)	73.8	0.010	Ref		
MSS	233 (80)	55.7		2.02	1.03–3.95	0.040
Sex						
Female	156 (54)	60.5	0.677	Ref		
Male	135 (46)	57.7		1.06	0.72–1.56	0.782
Age						
<60	46 (16)	79.9	<0.004	Ref		
60–70	53 (18)	65.4		1.91	0.84–4.32	0.122
70–80	118 (41)	53.9		2.91	1.42–5.97	0.004
>80	74 (25)	50.3		3.15	1.48–6.73	0.003
Tumor location						
Proximal colon	133 (46)	64.9	0.010	Ref		
Distal colon	91 (31)	58.1		1.18	0.73–1.91	0.505
Rectum	67 (23)	49.5		2.23	1.33–3.74	0.002
pT stage						
3	272 (93)	59.6	0.458	Ref		
4	19 (7)	52.6		1.72	0.84–3.50	0.138
Histopathologic grade						
G1/G2	250 (86)	59.3	0.756	Ref		
G3	32 (11)	62.8		1.61	0.79–3.30	0.190
Mucinous	6 (2)	66.7		1.41	0.32–6.17	0.647
Surgery						
Elective	252 (87)	61.2	0.018	Ref		
Acute	39 (13)	45.7		1.81	1.07–3.08	0.028

[a]Kaplan–Meier estimate, log-rank test.
[b]Cox regression, all included variables are displayed in the table.

camouflage an otherwise better prognosis for MSI tumors in stage III in our series. The patients who have received adjuvant treatment comprise 56 patients of whom 11 had MSI tumors. Excluding these from the analyses did not result in increased prognostic impact of MSI in stage III (data not shown).

The clinical applicability of MSI as a prognostic marker remains to be decided. Clearly, stage II tumors in the proximal colon make up the interesting subgroup because of the high prevalence of MSI (38%). Stage II patients do not routinely receive adjuvant therapy according to Norwegian guidelines. This seems reasonable for patients with an expected 5-year relative survival of 75% [12]. However, the MSS subgroup of patients had significantly worse prognosis, and these patients might benefit from adjuvant therapy. To demonstrate such a benefit, a randomized trial is necessary. Additional molecular markers may refine the poor and good MSI-based prognostic groups such as the recent ColoGuideEx, a 13 gene expression signature specific to stage II patients published by our group [38].

The prevalence of MSI in the current series was 14%. This is in accordance with comparable series [33, 39–42]. The previous documented association of MSI phenotype with right-sided colorectal cancer was confirmed. MSI was also more common in women than in men, partly due to the fact that women had a higher proportion of their tumors in the proximal colon (49%) compared with men (31%), which is in agreement with a study from New Zealand [43], but also because women had a higher frequency of MSI in their proximal tumors than men.

We found no significant association between MSI status and age. Other studies report the highest frequencies of MSI tumors in the oldest patients [28, 33, 44].

The proportion of MSI tumors was highest in stage II. This observation is in compliance with several other studies [24–26, 28, 40, 42]. The low number of MSI tumors in stage I in the present series can partly be explained by few stage I tumors in the proximal colon and numerous stage I tumors in the rectum. This finding might be connected to the absence of systematic screening for colorectal cancer in Norway which implies that most patients have developed symptoms at the time of diagnosis. Tumors in the proximal colon typically cause more subtle symptoms than tumors in the distal colon and rectum and may have reached a more advanced stage by the time of detection. The high frequency of MSI in stage II tumors might also reflect a less aggressive phenotype with lower tendency to metastasize [25].

The number of examined lymph nodes was low in this series, but probably representative for consecutive series from a

routine setting in this period. However, the low number should not introduce any bias in the calculations since this influences MSI/MSS and different tumor locations equally. Other authors have reported a higher number of examined lymph nodes in MSI patients [45–47], and suggested that MSI tumors induce larger lymph nodes which are more easily identified and retrieved by the pathologist. However, when adjusting for tumor location, the effect of MSI disappeared [47]. This is in line with our finding. A probable explanation is that different tumor locations result in different anatomical resections with unequal numbers of lymph nodes due to the anatomical distribution of mesocolic lymph nodes.

There is a correlation between the number of examined lymph nodes and correct staging [9], and this might explain why stage III patients have the highest number of examined lymph nodes. The correlation between the number of examined lymph nodes and age has also been described by others [10]. In the present series, a higher proportion of patients <60 years in the more recent years, corresponding to a period with increasing number of examined lymph nodes [48], might explain this.

In conclusion, the present study demonstrates that MSI is a positive prognostic factor in patients with stage II colon cancer, but not in stage III. MSS could be a clinical useful biomarker for the identification of patients with stage II right-sided colon cancer at increased risk of relapse.

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disclosure

The authors have declared no conflicts of interest.

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VEGFR-2, CXCR-2 and PAR-1 germline polymorphisms as predictors of survival in pancreatic carcinoma

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Background: Hypoxic environment of pancreatic cancer (PC) implicates high vascular in-growth, which may be influenced by angiogenesis-related germline polymorphisms. Our purpose was to evaluate polymorphisms of vascular endothelial growth factor receptor 2 (VEGFR-2), CXC chemokine receptor 2 (CXCR-2), proteinase-activated receptor 1 (PAR-1) and endostatin (ES) as prognostic markers for disease-free (DFS) and overall survival (OS) in PC.
Patients and methods: Genotyping of 173 patients, surgically treated for PC between 2004 and 2011, was carried out by TaqMan® genotyping assays or polymerase chain reaction. Chi-square test, Kaplan–Meier estimator and Cox regression hazard model were used to assess the prognostic value of selected polymorphisms.
Results: VEGFR-2 −906 T/T and PAR-1 −506 Del/Del genotypes predicted longer DFS ($P = 0.003$, $P = 0.014$) and OS (VEGFR-2 −906, $P = 0.011$). CXCR-2 +1208 T/T genotype was a negative predictor for DFS ($P < 0.0001$). Combined analysis for DFS and OS indicated that patients with the fewest number of favorable genotypes simultaneously present (VEGFR-2 −906 T/T, CXCR-2 +1208 C/T or C/C and PAR-1 −506 Del/Del) were at the highest risk for recurrence or death ($P < 0.0001$).

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