Susceptibility of *Msh2*-deficient Mice to Inflammation-associated Colorectal Tumors

Maija R. J. Kohonen-Corish, Joseph J. Daniel, Hein te Riele, Gary D. Buffinton, and Jane E. Dahlstrom

Department of Medicine, University of Sydney, Sydney, NSW 2006 & Division of Molecular Medicine, John Curtin School of Medical Research, Canberra, ACT 0200, Australia; [M. R. J. K-C.]; Department of Molecular and Clinical Genetics, Royal Prince Alfred Hospital, Sydney, NSW 2050, Australia [J. J. D.]; The Netherlands Cancer Institute, 1066 cx Amsterdam, the Netherlands [H. t. R.]; and The Canberra Clinical School, University of Sydney [G. D. B., J. E. D.] and ACT Pathology [J. E. D.], The Canberra Hospital, Canberra, ACT 2605, Australia

ABSTRACT

Patients with longstanding extensive ulcerative colitis have an increased risk of developing colorectal cancer (CRC). There are significant differences in the early pathogenesis of colitis-associated tumors compared with common CRC, whereas the frequency, degree, and significance of microsatellite instability (MSI) as a marker of mismatch repair deficiency in colitis tumors remain unclear. Here we describe the application of the DSS model of chronic colitis to mice with a defect in the *Msh2* mismatch repair gene to discern these early events. These mice do not develop CRC spontaneously without an external trigger. The aim of this study was to determine the effect of the *Msh2* defect on the frequency and grade of colitis-associated colorectal dysplasia and adenocarcinoma in *Msh2*+/−, *Msh2*+/−, and wild-type (*Msh2*+/+) mice and on the MSI status of the tumors. We show that in mice with chronic colitis, 60% of the *Msh2*+/− and 29% of the wild-type mice developed high-grade dysplasia or adenocarcinoma, but heterozygosity for the *Msh2* defect did not increase tumor susceptibility over wild-type genotype. The largest difference between genotypes was in the frequency of high-grade dysplasia, with 46.7, 8, and 12.5% in *Msh2*+/−, *Msh2*+/−, and *Msh2*+/+ mice, respectively. The *Msh2*+/− mice developed MSI-high tumors, whereas the majority of the *Msh2*+/− and wild-type tumors had no MSI. In the *Msh2*+/− mice, MSI appeared early in non-neoplastic colon tissue, presumably as a result of markedly increased epithelial cell proliferation associated with inflammation. These observations suggest that a homozygous mismatch repair defect predisposes to tumors triggered by chronic inflammation but is not the only factor involved because tumors also developed in the wild-type mice. This model of colitis offers opportunities to characterize the different molecular pathways of carcinogenesis operating in chronic colitis.

INTRODUCTION

Colorectal cancer is one of the most serious and frequent complications of long-standing IBD, particularly in chronic ulcerative colitis, which 10 years after onset, the cumulative risk is 0.5–1% per year in pancolitis patients (1). There are significant differences in the early pathogenesis and molecular alterations of ulcerative colitis-associated tumors as compared with common sporadic colorectal cancer implying that at least the early carcinogenesis is different (2). Dysplasia in sporadic cancers usually presents as a well-defined polyp, whereas dysplasia in ulcerative colitis is diffuse and macroscopically flat. It is well known that failure of cellular mechanisms to detect and repair defects in the DNA sequence is associated with carcinogenesis. DNA-free radical species, oxidative injury, and depletion of antioxidants in the inflamed mucosa (13–15) similar to that in human IBD (16, 17). Initially, DSS produces an acute colitis, which becomes chronic after repeated cycles of DSS/water. DSS is not a carcinogen on its own, but dysplasia and adenocarcinoma develop in a small subset of mice with chronic colitis (18).

The *Msh2*-deficient mice have a defect in one of the DNA mismatch repair genes that in humans cause susceptibility to HNPCC (19–21). However, there are some important differences. Mice homozygous for the *Msh2* defect (*Msh2*+/−) develop late-onset adenocarcinoma in the small intestine rather than in the colon, the latter of which is the main site of cancer in HNPCC. Mice heterozygous for the *Msh2* defect do not develop spontaneous small intestinal or colon tumors in contrast to heterozygous carriers of the corresponding human mutations. The aim of this study was to examine the effect of chronic inflammation on the frequency of colorectal dysplasia and cancer in *Msh2*-deficient mice and examine the MSI status of the tumors. We show that *Msh2*+/− mice develop a significantly higher frequency of colon tumors than the heterozygous *Msh2*+/− and wild-type *Msh2*+/+ mice. The MSI status of the lesions was also different, with the *Msh2*+/− mice developing tumors with MSI, whereas the heterozygous and wild-type mice developed mostly MSS neoplasia. This system offers opportunities to characterize the different molecular pathways of carcinogenesis operating in chronic colitis.

MATERIALS AND METHODS

Mouse Breeding. The *Msh2*+/− mice on the 129/Ola background (20) were crossed with wild-type C57BL/6J to obtain *Msh2*+/− males and females. These were subsequently crossed to produce *Msh2*+/−, *Msh2*+/−, and *Msh2*+/− litters. The *Msh2* genotype was determined by using a 3-primer PCR from mouse ear punch tissue developed by N. Toft (University of Edinburgh, Molecular Medicine Center, United Kingdom). The primers were P1 5′-CGG CCT TGA GCT AAG TCT ATT ATA AGG; P2 5′-GTT GGG ATT AGA

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The abbreviations used are: IBD, inflammatory bowel disease; MSI, microsatellite instability; MSH2, microsatellite instability-high; MSS, microsatellite instability-low; MSS, microsatellite stable; DSS, dextran sodium sulfate; HNPCC, hereditary nonpolyposis colorectal cancer; TGFβRII, transforming growth factor β receptor type II.
Induction of Chronic Colitis with DSS. Groups of Msh2+/−, Msh2−/−, and Msh2−/− mice (12–15 week old) were given repeated cycles of 5% DSS in drinking water as described previously to induce chronic colitis (11) followed by distilled drinking water for 2 weeks. The control genotypes received distilled water only. DSS was normally given for 4 days except for 3 days in cycle 2 (males). The females were given an extra 2 days of DSS in the first cycle to boost the onset of colitis symptoms to the same level as the males. The consumption of DSS solution per cage (3–5 mice) was measured throughout the experiment, and the fluid was changed daily. The mice were also weighed daily, and the symptoms of diarrhea and bleeding were recorded individually for each mouse. Symptoms were scored as either absent (0), slight (1), or severe (4) to permit a qualitative, noninvasive assessment of disease severity. If excessive weight loss or other excessive symptoms were observed, mice were sacrificed early. Otherwise, mice were killed, and organs were collected after five or eight cycles of DSS/water. This study was approved by the Animal Experimentation Ethics Committee at the Australian National University.

Histopathology. A total of 162 mice (90 females and 72 males) was analyzed for histopathology. After death, the entire small and large intestines (including anal verge) were removed, opened longitudinally, cleaned, examined for abnormalities, and placed in 10% neutral buffered formalin. After 24 h, these were transferred to 70% ethanol until processed for histopathology. The whole colon was embedded into three paraffin blocks (divided into distal, transverse, and proximal sections, approximately five pieces of tissue per block). Multiple sections of each block were examined. The stomach and representative pieces of the small intestine were mounted for analysis. All blocks, including untreated controls, were analyzed by an anatomical pathologist without knowledge of the treatment given. The features assessed included: inflammation (acute or chronic and graded as mild, moderate, or severe) and presence or absence of ulceration, architectural distortion and crypt loss, lymphoid aggregates, paneth cell metaplasia, and epithelial dysplasia or malignancy. Dysplasia was graded as negative, indefinite for low-grade dysplasia and low- and high-grade dysplasia according to the Riddell classification (22). Adenocarcinomas were classified as mucinous if they showed >50% mucinous differentiation. Presence or absence of lymphomas in the large and small intestine was also recorded. Results were scored on pathology sheets, then decoded and arranged according to genotype. All of the blocks that were classified as indefinite dysplasia had additional multiple levels examined and reanalyzed for pathology. Dysplastic and cancerous areas were marked on each slide for subsequent MSI and mutation analysis.

MSI Analysis. Areas of dysplasia and adenocarcinoma were microdissected from H&E-stained serial sections. Normal tissue from the stomach or ear punch DNA from the same mouse was used as a control for each tumor. DNA was prepared using the Puregene DNA Isolation Kit (Gentra Systems, Minneapolis, MN). One mononucleotide (urokinase receptor gene, uPAR) and three dinucleotide markers (D1Mit62, D15Mit93, and D17Mit72) were selected from previous studies (20, 23). The primer sequences for the uPAR marker were kindly supplied by Dr W. Edelmann, 5′-GCT CAT CTT CGT TCC CGT TC (forward strand) and 5′-CAT GTG GAA AGC TCT GA (reverse strand). In addition, a T20 repeat in the mouse pro-1 gene (GenBank M29169) was tested using primers 5′-AGC CAT CAA ACT GAT TTC CA (forward strand) and 5′-ACA AAG GGA GTG CTA GGT CA (reverse strand). This panel of two mononucleotide and three dinucleotide markers was used to determine the degree of MSI following the recommendation for human colorectal cancer (5). Tumors were classified MSI-H if two or more of the five markers showed instability or MSI-L if only one marker was unstable. An additional two markers, D17Mit123 and D17Mit185, were studied in MSI-L and MSS specimens to confirm MSI status. Primer sequences for all dinucleotide markers were obtained from the Whitehead Institute/MIT Center for Genome Research database.4

One primer from each primer pair was synthesized with a 5′ fluorescent tag (HEX, FAM, or TET). PCR amplification was performed in a total volume of 20 μl containing 25–100 ng of DNA, 250 μM deoxynucleotide triphosphates, 35–55 ng of each primer pair, and 1.2 units of Taq polymerase (Applied Biosystems, Foster City, CA). PCR was carried out at 94°C (30 s), 56°C–60°C (30 s), and 72°C (30 s) for 35 cycles, after an initial denaturation step of 2 min and with a final elongation step of 60 min. PCR was carried out with the GeneAmp PCR system 9600 and 2400 (Applied Biosystems). Fragments were visualized using ABI 310 Genetic Analyzer (Applied Biosystems) capillary electrophoresis. Specimens were scored as having MSI if there was a significant shift in the microsatellite pattern compared with matching normal tissue which produced a consistent pattern of peaks. The highest peak was chosen as the reference. If this fragment amplified less strongly (i.e., produced a lower peak) than at least one of the neighboring fragments in tumor tissue, this was classified as MSI. In addition, extra peaks often appeared that were negligible or not present in normal tissue.

Analysis of Short Mononucleotide Repeats. Three short mononucleotide repeats, JH102, JH104 (24), and a A5G6 repeat, from the TGFβRII (25), were analyzed for insertion-deletion mutations in the dysplasia and cancer specimens. The primer sequences for TGFβRII were 5′-CAT TAC TCT GGA GAC GGT TTG C (forward strand) and 5′-TGG TTG CAC TCT TCC ATG TTA C (reverse strand). PCR was carried out with fluorescent primers as above, except that the annealing temperature was 56°C with a 7-min final extension. Fragments were visualized using the ABI 310 Genetic Analyzer. The JH104 and the TGFβRII fragments were also sequenced to confirm results obtained from PCR fragment length analysis.

Statistical Analysis. The effect of the Msh2 genotype on the frequency of dysplasia and carcinoma was analyzed by χ2 with exact probability (program StatXact; Cytel Corp., Cambridge, MA). The frequencies were subdivided into four categories (normal, low-grade dysplasia, high-grade dysplasia, and adenocarcinoma), and pair-wise comparisons were made between the different genotypes. Daily basal water consumption (mean/mouse) was determined and analyzed during the first two cycles of DSS supplementation by general linear model ANOVA with genotype and sex as categorical variables (program SYSTAT 7.0.1; SPSS, Inc., Chicago, IL).

RESULTS

Induction of Chronic Colitis. Groups of Msh2+/+, Msh2−/−, and Msh2−/− mice were given repeated cycles of DSS/water, which caused chronic diarrhea and occasional rectal bleeding. The mean daily consumption of DSS-supplemented water did not differ significantly between genotypes (P = 0.39, general linear model ANOVA), but males consumed more than females (P < 0.001) as expected according to their higher body weight. This was compensated by giving the females 1–2 extra days of DSS in the first two cycles. The least-square means were 3.85, 3.78, and 4 ml/day for the three genotypes +/+ , +/−, and −/−, respectively. Nine DSS-treated Msh2−/− mice were sacrificed after three cycles of DSS, because of severe symptoms of colitis or bowel obstruction (7 mice) or large thymic lymphoma (2 mice). Eight of 9 Msh2−/− mice had developed dysplasia or cancer (4 low-grade, 3 high-grade dysplasias, and 1 adenocarcinoma). Six Msh2+/+ and 2 Msh2−/− mice also had to be sacrificed at this time point because of severe colitis symptoms, but dysplasia had developed in only 2 Msh2+/− mice (one low grade and one high grade) and 1 Msh2−/− mouse (high grade). The rest of the mice were sacrificed after five or eight cycles of DSS.

Microscopically, mice treated with DSS, showed diffuse and often transmural chronic active inflammation of the colon (Fig. 1B). The inflammatory changes were more marked distally. The colitis was characterized by crypt distortion and crypt loss, basal plasmacytosis, cryptitis, focal crypt abscess formation, accumulation of foamy macrophages, and lymphoid aggregates. There was often squamous metaplasia of the rectum. Paneth cell metaplasia was not a feature of the inflammatory changes. There was no significant small bowel inflammation. The severity of inflammation was similar in all three genotypes with ~80–90% of DSS-treated mice showing symptoms of severe chronic inflammation.
Colorectal Dysplasia and Cancer. Most (28 of 30) $Msh2^{-/-}$ mice developed some degree of dysplasia or cancer. Almost half (46.7%) of the $Msh2^{-/-}$ mice developed high-grade dysplasia (Table 1 and Fig. 1C), whereas only 12.5% of the $Msh2^{+/-}$ ($P = 0.002, \chi^2$) and 8% of the $Msh2^{+/-}$ mice had this lesion ($P = 0.0001$). After eight cycles, the proportion of adenocarcinoma increased in both the $Msh2^{+/-}$ and $Msh^{+/-}$ mice, but the overall frequency of neoplasia was still 35 and 46%, respectively. About 81% of adenocarcinomas showed mucinous differentiation (Fig. 1D), which is also a feature of human ulcerative colitis. Adenocarcinoma was associated with low- or high-grade dysplasia in 6 of 12 $Msh2^{+/-}$ mice, 9 of 13 $Msh2^{+/-}$ mice, and 3 of 4 $Msh2^{+/-}$ mice. Table 1 summarizes the pathology results for each genotype on dysplasia/carcinoma development is statistically significant after three to five cycles of DSS/water ($Msh2^{-/-}$ vs. $Msh2^{+/-}$, $P = 0.002, \chi^2$; $Msh2^{-/-}$ vs. $Msh2^{+/-}$, $P = 0.0001$; $Msh2^{+/-}$ vs. $Msh2^{+/-}$, $P = 0.69$).

Table 1 Percentage of mice that developed dysplasia/cancer in the colon with DSS treatment

<table>
<thead>
<tr>
<th>Genotype</th>
<th>No treatment</th>
<th>DSS</th>
<th>DSS</th>
<th>DSS</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Neoplasia</td>
<td>Indef + LGD</td>
<td>HGD</td>
<td>AdCa</td>
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<tr>
<td></td>
<td>% (n)</td>
<td>% (n)</td>
<td>% (n)</td>
<td>% (n)</td>
</tr>
<tr>
<td>3–5 cycles of DSSa</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$Msh2^{+/-}$</td>
<td>0 (0/10)</td>
<td>25.0 (6/24)</td>
<td>12.5 (3/24)</td>
<td>16.7 (4/24)</td>
</tr>
<tr>
<td>$Msh2^{-/-}$</td>
<td>33.3 (10/30)</td>
<td>46.7 (14/30)</td>
<td>13.3 (4/30)</td>
<td></td>
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<tr>
<td>8 cycles of DSSb</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>$Msh2^{+/-}$</td>
<td>0 (0/28)</td>
<td>8.1 (3/37)</td>
<td>5.4 (2/37)</td>
<td>21.6 (8/37)</td>
</tr>
<tr>
<td>$Msh2^{-/-}$</td>
<td>15.2 (7/46)</td>
<td>6.5 (3/46)</td>
<td>23.9 (11/46)</td>
<td></td>
</tr>
</tbody>
</table>

a Effect of the $Msh2^{-/-}$ genotype on dysplasia/carcinoma development is statistically significant after three to five cycles of DSS/water ($Msh2^{-/-}$ vs. $Msh2^{+/-}$, $P = 0.002, \chi^2$; $Msh2^{-/-}$ vs. $Msh2^{+/-}$, $P = 0.0001$; $Msh2^{+/-}$ vs. $Msh2^{+/-}$, $P = 0.69$).

b Indefinite, indefinite; LGD, low grade; HGD, high-grade dysplasia; AdCa, adenocarcinoma. The frequency of indefinite and low-grade dysplasia is combined because of the occasional difficulty in differentiating between the two (22).

c $P = 0.77$.

Microsatellite Analysis. Lymphomas and intestinal tumors from the Msh2\(^{-/-}\) mice are known to display MSI (19–21, 26). We first tested 20 of these spontaneous tumors from the Msh2\(^{-/-}\) mice with markers D1Mit62, D15Mit93, D17Mit72, uPAR, and pro-1 and observed MSI in all specimens with one or more of the markers. Colorectal dysplasias and carcinomas from 83 DSS-treated mice were then analyzed for MSI (Table 2). Both the inflammation-associated colorectal tumors and the spontaneous intestinal adenocarcinomas from the Msh2\(^{-/-}\) mice displayed subtle changes from the reference microsatellite patterns (Fig. 2, A and B). The most sensitive indicator of MSI was the T\(_{24}\) tract in an intron of the uPAR gene. Altogether, 21 of 27 Msh2\(^{-/-}\) colorectal tumors showed instability in this locus and in at least one other marker (Fig. 2B). Therefore, these tumors were classified MSI-H following the standard criteria for human colorectal cancer (5). Three of the 27 Msh2\(^{-/-}\) colorectal tumors showed instability in only one marker (MSI-L), and three were MSS. Other areas of the intestinal tract, such as stomach and proximal small intestine in the DSS-treated mice, showed the same pattern as the normal colon in untreated Msh2\(^{-/-}\) mice (Fig. 2D). The Msh2\(^{-/-}\) tumors analyzed included three adenocarcinomas and 14 high-grade, 8 low-grade, and 2 indefinite dysplasias (Table 2). Of these, 1 high-grade and 2 low-grade dysplasias were MSI-L, and 2 low-grade and 1 indefinite dysplasia were MSS. There was no correlation between the MSI status and the location of the tumor in the colon.

Tumors from 3 Msh2\(^{+/+}\) and 4 Msh2\(^{-/-}\) mice were MSI-L, and none showed instability in the uPAR marker. All MSI-L and MSS tumors were also analyzed with a further two dinucleotide markers, D17Mit123 and D17Mit185, but these results did not change the MSI status of any sample. The Msh2\(^{-/-}\) tumors that were MSI-L were also studied for loss of heterozygosity using the Msh2 locus-specific PCR, but both the wild-type (Msh2\(^{+}\)) and the targeted (Msh2\(^{-}\)) alleles were retained in these samples (data not shown).

Regions of colon with no identifiable dysplasia or cancer were selected for additional MSI analysis from 22 DSS-treated Msh2\(^{-/-}\) mice that had developed tumors. All tissue sections surrounding the identified tumor in the block were analyzed and included both inflamed and normal tissue. MSI was evident in 15 of 22 specimens, particularly with the mononucleotide markers (Fig. 2C). This implies that some MSI is induced by colon inflammation and can precede dysplasia in Msh2\(^{-/-}\) mice. The exceptions included 2 mice whose tumors were MSS and 5 mice whose tumors were MSI-H.

Mutations in Short Mononucleotide Repeats and the TGFβRII Gene. To examine whether the MSI observed would also cause deletion-insertion mutation in shorter repeat sequences, an additional three genes were analyzed. The JH102 and JH104 repeats have been reported previously to be good indicators of MSI in tumors of Mlh1-deficient mice (24). JH102 is an A\(_{10}\) repeat in the coding region of the mouse Apc gene. JH104 is located in an intron of the Msh3 DNA mismatch repair gene. No insertion-deletion mutations were found in any of these markers in 10 MSI-H Msh2\(^{-/-}\) colorectal tumors analyzed. The JH104 marker was also found to be polymorphic, with a T\(_9\) repeat in the C57BL/6J strain and a T\(_{10}\) repeat in the 129/OLA strain. The A\(_{10}\)GA\(_4\) repeat in the coding region of the TGFβRII gene corresponds to the A\(_{10}\) repeat known to be mutated in human MSI-H colorectal cancers (27) but has not been studied previously in intestinal or colorectal tumors in these mice. We could not find any evidence of insertion-deletion mutations of the A\(_{10}\)GA\(_4\) repeat in colorectal tumor specimens from 21 DSS-treated Msh2\(^{-/-}\) mice.

DISCUSSION

The recognition of DNA mismatch repair genes in hereditary and common colorectal cancer (3, 4) has led to the analysis of the role of these genes in IBD-associated cancer (6, 7). Genetic association studies implicated mismatch repair genes as minor susceptibility genes when the frequencies of an MSH2 splice site substitution were compared in two groups of patients with ulcerative colitis, one with high-grade dysplasia and cancer and one without tumors (28). It was noted that this substitution was more common in the patient group with tumors and concluded that a variant of MSH2, which is not normally deleterious, may predispose to cancer in the setting of chronic colitis. A later study (29) was unable to confirm the association of colitis cancer with MSH2. A significant association between Crohn’s disease/ulcerative colitis, certain polymorphic alleles of the MLH1 gene, and a microsatellite marker located in an intron of MLH1 has been reported (30). However, there is no association between HNPCC and an increased risk for IBD.

This study describes the effect of a DNA mismatch repair defect on the development of dysplasia and cancer when triggered by chronic inflammation. We analyzed mice that were either homozygous, heterozygous, or wild type for the Msh2 DNA mismatch repair defect.
There was no difference in the severity of inflammation between the three genotypes. Heterozygosity for the Msh2 defect did not increase tumor susceptibility in the setting of chronic inflammation compared with wild type. Heterozygous carriers of a MSH2 gene defect in humans ultimately develop HNPCC from their late teens as the wild-type allele is mutated in colon epithelial cells. It is not known what triggers the somatic mutation in HNPCC, and no data are available from human IBD patients who also are MSH2 mutation carriers. The majority of the Msh2+/− and wild-type tumors in our study appeared to develop independently of the mismatch repair system as very little MSI was found.

In contrast to the maximum frequency of ~30% of high-grade dysplasia or adenocarcinoma found in the wild-type or Msh2+/− mice, 60% of the DSS-treated Msh2−/− mice developed these tumors. The largest difference was observed in the frequency of high-grade dysplasia, with 12.5, 8, and 46.7% in Msh2+/−, Msh2−/−, and Msh2−/− mice, respectively, whereas the frequency of adenocarcinoma was similar for each genotype. These results suggest an important connection between the DNA mismatch repair proteins and development of dysplasia and contrast a widely held view that mismatch repair defects accelerate carcinogenesis (31, 32). There is increasing evidence that the mismatch repair machinery is not only activated by mismatched nucleotides but also by DNA damaging agents (33, 34). It has been suggested that the mismatch repair proteins may signal damage-induced apoptosis through a direct-sensor signaling process (32, 35). Our study suggests that inflammation-induced damage may also be controlled by mismatch repair proteins, such as MSH2. It will be important to determine whether chronic colitis involves a similar apoptotic pathway (32, 33).

We also observed a significant difference in the frequency of MSI, with the majority of Msh2−/− tumors being MSI-H. However, the individual MSI patterns were much more subtle than we have observed in human colorectal cancer specimens using similar methods but no different from the spontaneous intestinal cancers from the same mice. Nevertheless, the tumor MSI patterns were absolutely reproducible and quantifiable. Interestingly, we were not able to detect any instability in short mononucleotide repeats, which have been reported previously to be mutated in tumors from mismatch-deficient mice. These included the A2GA4 repeat in the coding region of the TGFBR II gene, as reported to be mutated in Msh2−/− lymphomas (25). The corresponding A10 repeat is also an important mutation target in human MSI-H colorectal cancers (27). This indicates that other target genes may be mutated or abnormally regulated in this model.

In the majority of DSS-treated Msh2−/− mice tested, MSI was also detected in the colon tissue surrounding the tumor, implying that some MSI is already induced by inflammation and precedes dysplasia. Instability at one or more microsatellite loci has also been found in a proportion of human inflamed non-neoplastic lesions in IBD patients (28, 29, 36). It has been suggested that this is caused by saturation of normal DNA repair mechanisms in inflamed colon epithelium because of markedly increased cell turnover and proliferation in IBD (29). Our results are compatible with the hypothesis that the increased proliferation induces widespread DNA replication errors, which are repaired normally until the DNA repair systems become saturated. As a result of defective mismatch repair already present in the Msh2+/− mice, this is quickly manifested by the appearance of MSI in the inflamed colon tissue. In the wild-type and Msh2+/− mice, chronic inflammation causes the same DNA damage, but the intact mismatch repair system is able to repair more of the damage.

This still leaves a high proportion of MSS dysplasia and cancer that cannot be explained on the basis of temporary saturation of the DNA mismatch repair system and probably includes the majority of human IBD cancers. Therefore, other mechanisms will need to be looked at, such as gene silencing by hypermethylation of CpG-rich promoters and injury to DNA bases directly as a result of the inflammatory response. In conclusion, we have described a novel application of a previously well-characterized animal model of chronic colitis. We have shown that by studying colorectal dysplasia and cancers developing in mice that are either homozygous, heterozygous, or wild type for a DNA mismatch repair defect, parallels can be drawn to human chronic colitis-associated cancers. Additional molecular genetic analysis of this model is likely to reveal more insights to the early pathogenesis of dysplasia and the different molecular mechanisms of carcinogenesis operating in IBD cancers.

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