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Cancer in Inflammatory Bowel Disease: lessons from animal models

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Abstract

Purpose of the review—Human colitis-associated cancers (CAC) represent a heterogeneous group of conditions in which multiple oncogenic pathways are involved. In this manuscript we reviewed the latest studies using genetic, chemically induced, bacterial and innate immunity induced experimental models of colitis-associated cancer.

Recent findings—Using the azoxymethane-dextran sodium sulfate model wound healing pathways seems to be required in the development of CAC. There is also an emerging understanding that commensal and/or pathogenic bacteria can promote tumorigenesis, through T cell mediated inflammation. Using specific transgenic mice (villin-CD98, T cell SMAD7, villin-TLR4) or specific knock-out mice, investigators have identified that derangements in epithelial or innate and adaptive immune pathways can result in CAC. Subtle perturbations in epithelial repair —both too little or too exuberant, can render mice susceptible to tumorigenesis.

Summary—With the aid of animal models, we have witnessed a rapid expansion of our knowledge of the molecular and immunologic mechanisms underlying inflammatory cancers. Though animal models have contributed a discrete amount of information to our understanding of tumorigenesis in the setting of intestinal inflammation it is clear that no single animal model will be able to adequately recapitulate the pathogenesis of complex CRCs, but each model gets us one step closer to comprehending the nature of CAC.

Keywords

mouse; AOM; DSS; colitis cancer; APC

Introduction

CACs are malignancies occurring in the setting of chronic inflammatory disorders of the colon. In humans, CAC is most commonly encountered in inflammatory bowel diseases (IBD), both ulcerative colitis (UC) and Crohn's disease. CAC may occur in as many as 18.4% of IBD patients over the course of 30 years [1]. Clinical inquiry has revealed a number of factors associated with the development of CAC in humans, including duration of IBD, extent of colonic involvement, severity of inflammation, and family history of colon cancer [2–6].

Despite an understanding of the clinical features associated with CAC, the molecular mechanisms underlying carcinogenesis in the setting of chronic colitis are unclear. What we

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do know regarding CAC in humans is gleaned from histopathologic observation and molecular studies. Chromosomal instability (CIN) is an early and frequent abnormality among patients with CAC, often associated with malfunction of tumor suppressor genes, shortening of telomeres, and aneuploidy [7–10]. Another form of genomic instability, microsatellite instability (MSI) is also frequently observed in the setting of CAC [11, 12]. Although observed early in sporadic (or non-inflammatory) colon cancers, mutations in the adenomatous polyposis coli (APC) gene are infrequent in dysplastic mucosa or neoplasias in the setting of colitis [13]. Yet, activated β-catenin is a frequent finding in colitis-associated dysplasia, and recent work has highlighted the role of Wnt/β-catenin signaling in maintenance of colon cancer stem cells [13–17].

Knowledge of the unique pathways of carcinogenesis underlying CAC in humans is currently inadequate. In order to clarify these pathways, a variety of animal models have been developed. This review will detail progress in rodent models of CAC.

Chemically induced colitis-associated neoplasia

Azoxymethane (AOM)- Dextran Sodium Sulfate (DSS) model

In laboratory rodents, colorectal cancer (CRC) is commonly induced with AOM, a potent carcinogen. Via alkylating species, DNA adducts form covalent bonds between the reactive carcinogen and the genomic DNA, resulting in mutations during DNA repair and promotion of tumor development. Chronic inflammation can also trigger neoplasia. Long-term administration or repeated cycles of DSS, a chemical that causes epithelial injury, can induce chronic colitis and subsequent dysplasia in rodents [18–20]. Currently the most used model of inflammatory CRC is the combination of AOM with DSS because tumor formation is enhanced compared to DSS alone [21–25]. As in human CAC, the degree of inflammation correlates with dysplasia in the AOM-DSS model and is associated with the nuclear translocation of β-catenin [26]. Up-regulation of mucosal Cox-2 and PGE_2 production are reported in the AOM-DSS model of colitis [27–29]. This model has been predominantly employed in mice, and its tumorigenic potential is strain dependent, as some genetic backgrounds such as A/J, BALB/c or Swiss Webster mice are more sensitive to AOM than other strains like C57Bl/6J mice [30–32].

More recent studies have attempted to further explain the pathogenic steps involved in the tumorigenic process of the rodent AOM-DSS model. Mice deficient in myeloid translocation gene related-1 (MTGR1), which display impaired wound healing, are resistant to CAC after AOM-DSS treatment despite having an active inflammatory infiltrate; this tumor resistance is due to increased intra-tumoral apoptosis [33]. This finding suggests that wound healing pathways are required in the development of CAC, and that wound healing and cancer exist on a continuum.

Other carcinogens

A variety of carcinogenic compounds have been used to experimentally reproduce the inflammatory events contributing to carcinogenesis. In classic studies, CAC was modeled in rats using carrageenan, a polysaccharide derived from red seaweed that has toxic effects, causing colonic ulceration and inducing tumorigenesis [34–36]. Other carcinogenic chemicals such as 1,2-dimethylhydrazine (DMH) [37], the precursor to AOM, or dietary iron [38], in combination with DSS, have been also used to chemically induce CAC. Additionally, 2-amino-1-methyl-6-phenylimidazol [4,5-b] pyridine (PhIP), a heterocyclic aromatic amine formed when cooking meat at a high temperature, has also been translated into experimental CRC. Rats fed with PhIP had a 50% incidence of colonic tumors [39]. Moreover, PhIP combined with a high fat diet or AOM enhances tumorigenesis [40]. A recent study suggested that glucagon-like peptide-2 (GLP-2), an intestinal growth factor secreted by

enteroendocrine cells, might be a colon cancer promoter in rats treated with PhIP combined with a high fat diet [41].

Genetic models of colitis and cancer

Genetic models of cancer in IBD have been developed to mimic known mechanisms underlying colitis and CACs in humans. These models have focused on mimicking immune dysregulation, inflammation, and epithelial barrier dysfunction as seen in human disease. Major advances have been made in this field with respect to genetic manipulation of APC gene [42, 43], p53 [44, 45], and IL-10 [46], among others [47–49]. Recent work in this field has built upon these findings, modifying conventional models or creating novel rodent paradigms. Table 1 provides an overview of the latest advances.

The development of mice deficient in APC gene was among the first successful applications of genetic modeling of intestinal cancers in rodents [50–52]. The superimposition of AOM or DSS to this model mimics CAC and results in more intestinal tumors [43]. Commensal microorganisms can also drive the development of colonic inflammation and colonic tumors in mice. For example, enterotoxigenic *Bacteroides fragilis*-mediated colitis in Min ($Apc^{+/-}$) mice results in the formation of colonic tumors via the inflammatory T_H17-dependent pathway [53]. IL-17A-secreting T cells were investigated by Chae et al. who looked at tumorigenesis, pro-inflammatory cytokines and immune abnormalities in $APC^{Min/+}$ and IL-$17A^{-/-}$ × $APC^{Min/+}$ mice. IL17-A ablation showed decreases in intestinal tumorigenesis and lower levels of IL-6, IL-23 and IL-1β, implicating these mediators in inflammatory cancers [54].

Many inflammatory colon cancers in humans also harbor genomic changes in the form of MSI that result from epigenetic deficiency in regulatory mismatch repair (MMR) proteins. These neoplasias can be flat in appearance, occur more often in the proximal colon, and histologically have mucin associated with a “Crohn’s-like” reaction. The deletion of the subunit protein Giα2 in mice causes growth retardation, diffuse spontaneous colitis resembling UC and non-polyposis right-sided, multifocal colorectal cancers arising from flat dysplastic lesions with many histologic features analogous to human dysplasia. At a young age, Giα2 knockout (KO) mice have normal colonic epithelium with a normal expression of MMR. In the setting of inflammation, however, older mice lose expression of the MMR proteins PMS2 and MLH1 in the inflamed colon tissue, cancer stroma and epithelium [55]. While epigenetic silencing by methylation of the MLH1 promoter is the abnormality identified in the majority of sporadic CRCs in humans [50, 51, 56], this was not seen in the Giα2 KO mice. Hypoxia causes suppression of MLH1 and PMS2 protein levels in colonic crypts by suppressing histone H3 acetylation at the proximal MLH1 promoter, leading to silencing of MLH1 and MSI [57].

To investigate the systemic effects of local immune dysregulation, mice with Giα2 KO and IL-10 deficiency have been created. IL-10 is produced by regulatory T cells (Tregs). Mice deficient in both Giα2 and IL-10 develop colitis at baseline; once treated with DSS, these mice acquire single and double-strand breaks in DNA of peripheral leukocytes; these systemic findings in response to local inflammation are thought to be related to the formation of systemic reactive oxygen and nitrogen species (RONS) [58]. Mice deficient in Nrf2 – a transcription factor involved in regulating oxidative stress - are more susceptible to chemically induced colitis with DSS compared to wild type mice and show higher rates of inflammation-associated neoplasia. These mice lose detoxifying enzymes and generate RONS [59–61]. These data suggest that colonic inflammation drives oxidative stress not only in the colon but also systemically, and that perturbations in the ability to modify production of RONS are linked to development of neoplasia.

Manipulation of IL-10 KO mice as a model for human IBD-colitis has helped to explain the role of the nucleotide-editing enzyme, activation-induced cytidine deaminase (AID), in inflammatory colon cancers. This enzyme induces somatic mutations in tumor-related genes, including p53 [62]. AID expression increases in inflammation compared to non-inflamed wild type mice. Mutations in p53 were more frequent in the IL-$10^{-/-}$ $AID^{+/+}$ model compared to the IL-$10^{-/-}$ $AID^{-/-}$, suggesting a role of AID in promoting mutations in the mouse model of CAC [63].

Transgenic mice have also been generated to study the role of immune dysregulation in CAC. In mice over-expressing Smad7 in T cells, a suppressor of TGF-β signaling, colitis increases in severity, but mice develop fewer colon tumors [64]. Increased expression of IFN-γ and increased accumulation of cytotoxic CD8+ and natural killer T cells were observed in these tumors and surrounding inflamed colonic tissue. Interestingly, in IFN$\gamma^{-/-}$xSmad7 transgenic mice, tumor incidence was restored to the same levels observed in WILD TYPE; thus, IFN-γ appears to play a major role in CAC.

Knock-out mice have also been generated to interrogate the role of wound healing in inflammation-related colon cancer development. Nod-like receptor pyrin domain-containing protein 6 (NLRP6) is primarily expressed by colonic myofibroblasts and plays a key role in mucosal wound healing. Following AOM-DSS administration, *Nlrp6*$^{-/-}$ mice had accelerated tumor growth and higher tumor burden than wild type mice, suggesting a role of NLRP6 in inflammation and carcinogenesis through an effect on myofibroblasts that help in epithelial repair [65].

The role of epithelial barrier dysfunction in CAC has also been explored with rodent models. CD98, a type II transmembrane glycoprotein adhesion molecule, transgenic mice have increased epithelial barrier dysfunction compared to wild type mice [66]. CD98 transgenic mice show more severe DSS colitis characterized by increased expression of the pro-inflammatory cytokines IL-1β and TNF-α. In the CD98 transgenic mice, AOM-DSS-induced colitis generates a larger quantity and size of tumors compared to wild type mice. TIR8 – a single immunoglobulin IL-1R-related molecule - inhibits signaling from the IL-1R/TLR complex and has inhibitory activity on inflammation. Mice deficient in TIR8 showed increased susceptibility to CAC when AOM was combined with DSS but not AOM alone, showing that inflammation was a prerequisite for carcinogenesis in this model [67]. TIR8 $^{-/-}$ mice display increased colon epithelial permeability compared with TIR8 $^{+/+}$ mice. In the TIR8 $^{-/-}$ mice, many pro-inflammatory mediators (e.g. IL-1β, IL-6, and TGFβ) are increased while IFN-γ is decreased [64].

Microbial models of CAC

The mammalian GI tract hosts a variety of commensal and symbiotic microorganisms that play an important role in intestinal homeostasis, maintenance, and function. Both the innate and adaptive immune systems of the host contribute to this homeostasis through the development of tolerance to commensal organisms while activating defense mechanisms against potential pathogens [68, 69]. It is theorized that the loss of this balanced interplay between the host and the microbiota contributes to the development of CAC. Changes in the intestinal flora of CAC patients have been reported. These studies based on 16S rRNA and high throughput sequencing reveal composition and diversity differences between the microbiota of CAC patients and that of healthy subjects; these differences have also been noted between chemical and genetic based murine models of CAC [70–73]. However, these comprehensive analyses of gut microbiota could not reveal whether this dysbiosis is the cause of CAC or an outcome of the mucosal abnormality.

Other studies based on gnotobiotic animals in conjunction with chemical and genetic models of CAC demonstrated that bacteria play a crucial role in the pathophysiology of CAC by promoting intestinal inflammation. Infection with enteric pathogens such as *Helicobacter* species and *Streptococcus bovis* have been clinically associated with CAC. Commensal bacteria are increasingly being used not only for understanding the mechanistic details of CAC but also to augment the AOM-induced response in animal models [74–76]. Both enterotoxigenic *Bacteroides fragilis* and *Citrobacter rodentium* promote colon tumor formation in $APC^{min/+}$ mice whereas the non-toxigenic strain of *B. fragilis* does not cause any tumor formation [77, 78]. The majority of the chemical and genetic models of CAC including IL-$2^{-/-}$, IL$10^{-/-}$, Gpx$1^{-/-}$, Tcr$\beta^{-/-}$ p$53^{-/-}$ mice under germ free conditions do not develop any tumors [79–81].

Certain bacterial species can also have a differential effect on the development of CAC, resulting in differing quantities, types and location of tumors. For example, germ-free IL-$10^{-/-}$ mice given either *E. faecalis* or *E. coli* yielded different clinical abnormalities, while more severe, earlier disease onset occurred when the mice were administered both bacteria simultaneously [82]. However, germ-free IL$10^{-/-}$ and IL$10^{-/-}$ mono-associated with other bacterial species including *Lactobacillus* and *Lactococcus* did not show any signs of CAC [83]. Gnotobiotic studies of TRUC mice (Tbet$^{-/-}$ × Rag$2^{-/-}$) dually associated with *Klebsiella pneumonia* and *Proteus mirabilis* did not induce colitis either; however, the addition of commensal organisms did result in colitis, indicating the necessity of both commensal and pathogenic organisms in inducing colitis [84].

Adoptive transfer models of CAC have also been studied. Adoptive transfer of Tregs from wild type mice into *H. hepaticus*-infected Rag$^{-/-}$ and $APC^{min/+}$ mice significantly reduced *H. hepaticus* induced CAC in these mice; this protective effect was more significant when Tregs were obtained from wild type mice that were previously infected with *H. hepaticus* [85, 86]. Based on these results, Erdman et al proposed a hypothesis whereby infections help in maintaining the anti-inflammatory phenotype of Tregs and reduce susceptibility to carcinogenesis [87].

In addition to identify pathogens or virulence factors that promote CRC, some investigators have tried to identify bacteria that protect against CAC. *Bifidobacterium lactis* protects against acute colitis and CAC in AOM-DSS treated mice and has been proposed as a possible therapy for CAC prevention [88]. The majority of the studies performed to date focus on the involvement of intestinal microbiota in CAC progression but clearly indicate that there is no single microbe or mechanism that can explain the underlying cause of CAC in humans.

Innate immune models of CAC

Another approach to model CAC in experimental animals is to target molecules involved in the innate immune response, which is the first stage of the intestinal inflammatory process. The innate immune receptors, including toll-like receptors (TLRs) and nucleotide-binding oligomerization domain receptors (NODs) have been used to study inflammation and cancer. Our group has developed a transgenic mouse that over-expresses TLR4 in the intestinal epithelium. These mice develop significantly more tumors than wild type mice after AOM-DSS treatment [89]. Moreover, we demonstrated that the majority of human patients with dysplasia and CAC over-express TLR4 in the intestinal epithelium. However, lacking TLR4 expression is also detrimental, as TLR4 deficient mice have worse colitis after DSS than wild type counterparts, but intriguingly are protected from AOM-DSS-induced CAC [89, 90]. Not all TLRs behave the same however, and Lowe et al. found that mice

deficient in TLR2 are prone to AOM-DSS tumorigenesis, showing a higher expression of inflammatory mediators such as IL-6, IL-17A, and STAT3 [91].

In the normal intestinal epithelium, TLRs are expressed in low levels. Inhibitors, like toll-interacting protein (TOLLIP) or single immunoglobulin IL-1-related receptor (SIGIRR), regulate TLR activity. Xiao et al reported that SIGIRR knockout mice are more susceptible to inflammation and CAC after AOM-DSS treatment. When SIGIRR was restored in the intestinal epithelium, CAC severity was decreased [92]. The myeloid differentiation primary response protein 88 (MyD88), an adaptor protein for TLR signaling pathways, also protects against AOM-DSS induced tumorigenesis. MyD88-deficient mice show impaired epithelial repair with decreased IL-18 signaling leading to increased inflammation and susceptibility to develop neoplastic lesions [93].

Conclusion

With the aid of animal models, we have witnessed a rapid expansion of our knowledge of the molecular and immunologic mechanisms underlying inflammatory cancers. Each of the models described above has contributed a discrete amount of information to our understanding of tumorigenesis in the setting of intestinal inflammation, and new genomic, immunologic, and gnotobiotic technologies offer promising possibilities. Yet, human inflammatory colorectal cancer is a heterogeneous disease with each tumor undergoing distinct molecular changes that accumulate over time. At present, it is clear that no single animal model will be able to adequately recapitulate the pathogenesis of complex CRCs, but each model gets us one step closer to comprehending the nature of CAC.

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References

1. Eaden JA, Abrams KR, Mayberry JF. The risk of colorectal cancer in ulcerative colitis: a meta-analysis. Gut. 2001; 48:526–535. [PubMed: 11247898]
2. Rutter MD, Saunders BP, Wilkinson KH, et al. Thirty-year analysis of a colonoscopic surveillance program for neoplasia in ulcerative colitis. Gastroenterology. 2006; 130:1030–1038. [PubMed: 16618396]
3. Ekbom A, Helmick C, Zack M, et al. Ulcerative colitis and colorectal cancer. A population-based study. N Engl J Med. 1990; 323:1228–1233. [PubMed: 2215606]
4. Rutter M, Saunders B, Wilkinson K, et al. Severity of inflammation is a risk factor for colorectal neoplasia in ulcerative colitis. Gastroenterology. 2004; 126:451–459. [PubMed: 14762782]
5. Gupta RB, Harpaz N, Itzkowitz S, et al. Histologic inflammation is a risk factor for progression to colorectal neoplasia in ulcerative colitis: a cohort study. Gastroenterology. 2007; 133:1099–1005. quiz 1340-1. [PubMed: 17919486]
6. Loftus EV Jr. Epidemiology and risk factors for colorectal dysplasia and cancer in ulcerative colitis. Gastroenterol Clin North Am. 2006; 35:517–531. [PubMed: 16952738]
7. Rabinovitch PS, Dziadon S, Brentnall TA, et al. Pancolonic chromosomal instability precedes dysplasia and cancer in ulcerative colitis. Cancer Res. 1999; 59:5148–5153. [PubMed: 10537290]
8. O'Sullivan JN, Bronner MP, Brentnall TA, et al. Chromosomal instability in ulcerative colitis is related to telomere shortening. Nat Genet. 2002; 32:280–284. [PubMed: 12355086]

9. Risques RA, Lai LA, Himmetoglu C, et al. Ulcerative colitis-associated colorectal cancer arises in a field of short telomeres, senescence, and inflammation. Cancer Res. 2011; 71:1669–1679. [PubMed: 21363920] * Study looking into the DNA and chromosomal changes in areas adjacent to high grade dysplasia and CAC in UC patients and the development from low grade dysplasia to colorectal cancer
10. Brentnall TA. Molecular underpinnings of cancer in ulcerative colitis. Curr Opin Gastroenterol. 2003; 19:64–68. [PubMed: 15699896]
11. Ishitsuka T, Kashiwagi H, Konishi F. Microsatellite instability in inflamed and neoplastic epithelium in ulcerative colitis. J Clin Pathol. 2001; 54:526–532. [PubMed: 11429424]
12. Tahara T, Inoue N, Hisamatsu T, et al. Clinical significance of microsatellite instability in the inflamed mucosa for the prediction of colonic neoplasms in patients with ulcerative colitis. J Gastroenterol Hepatol. 2005; 20:710–715. [PubMed: 15853983]
13. Aust DE, Terdiman JP, Willenbucher RF, et al. Altered distribution of beta-catenin, and its binding proteins E-cadherin and APC, in ulcerative colitis-related colorectal cancers. Mod Pathol. 2001; 14:29–39. [PubMed: 11211307]
14. van Dekken H, Wink JC, Vissers KJ, et al. Wnt pathway-related gene expression during malignant progression in ulcerative colitis. Acta Histochem. 2007; 109:266–272. [PubMed: 17445872]
15. Weber CR, Nalle SC, Tretiakova M, et al. Claudin-1 and claudin-2 expression is elevated in inflammatory bowel disease and may contribute to early neoplastic transformation. Lab Invest. 2008; 88:1110–1120. [PubMed: 18711353]
16. Mees ST, Mennigen R, Spieker T, et al. Expression of tight and adherens junction proteins in ulcerative colitis associated colorectal carcinoma: upregulation of claudin-1, claudin-3, claudin-4, and beta-catenin. Int J Colorectal Dis. 2009; 24:361–368. [PubMed: 19184060]
17. Sottoriva A, Verhoeff JJ, Borovski T, et al. Cancer stem cell tumor model reveals invasive morphology and increased phenotypical heterogeneity. Cancer research. 2010; 70:46–56. [PubMed: 20048071]
18. Okayasu I, Yamada M, Mikami T, et al. Dysplasia and carcinoma development in a repeated dextran sulfate sodium-induced colitis model. J Gastroenterol Hepatol. 2002; 17:1078–1083. [PubMed: 12201867]
19. Yamada M, Ohkusa T, Okayasu I. Occurrence of dysplasia and adenocarcinoma after experimental chronic ulcerative colitis in hamsters induced by dextran sulphate sodium. Gut. 1992; 33:1521–1527. [PubMed: 1333439]
20. Cooper HS, Murthy SN, Shah RS, et al. Clinicopathologic study of dextran sulfate sodium experimental murine colitis. Lab Invest. 1993; 69:238–249. [PubMed: 8350599]
21. Paulsen JE, Knutsen H, Olstorn HB, et al. Identification of flat dysplastic aberrant crypt foci in the colon of azoxymethane-treated A/J mice. Int J Cancer. 2006; 118:540–546. [PubMed: 16094649]
22. Suzuki R, Kohno H, Sugie S, et al. Dose-dependent promoting effect of dextran sodium sulfate on mouse colon carcinogenesis initiated with azoxymethane. Histol Histopathol. 2005; 20:483–492. [PubMed: 15736053]
23. Okayasu I, Ohkusa T, Kajiura K, et al. Promotion of colorectal neoplasia in experimental murine ulcerative colitis. Gut. 1996; 39:87–92. [PubMed: 8881816]
24. Suzuki R, Kohno H, Sugie S, et al. Strain differences in the susceptibility to azoxymethane and dextran sodium sulfate-induced colon carcinogenesis in mice. Carcinogenesis. 2006; 27:162–169. [PubMed: 16081511]
25. De Robertis M, Massi E, Poeta ML, et al. The AOM/DSS murine model for the study of colon carcinogenesis: From pathways to diagnosis and therapy studies. J Carcinog. 2011; 10:9. [PubMed: 21483655] ** Very good review going over all chemically induced colitis associated cancer models with a great emphasis in AOM/DSS
26. Cooper HS, Murthy S, Kido K, et al. Dysplasia and cancer in the dextran sulfate sodium mouse colitis model. Relevance to colitis-associated neoplasia in the human: a study of histopathology, B-catenin and p53 expression and the role of inflammation. Carcinogenesis. 2000; 21:757–768. [PubMed: 10753213]

27. Murakami A, Hayashi R, Tanaka T, et al. Suppression of dextran sodium sulfateinduced colitis in mice by zerumbone, a subtropical ginger sesquiterpene, and nimesulide: separately and in combination. Biochem Pharmacol. 2003; 66:1253–1261. [PubMed: 14505804]
28. Katayama K, Wada K, Nakajima A, et al. A novel PPAR gamma gene therapy to control inflammation associated with inflammatory bowel disease in a murine model. Gastroenterology. 2003; 124:1315–1324. [PubMed: 12730872]
29. Morteau O, Morham SG, Sellon R, et al. Impaired mucosal defense to acute colonic injury in mice lacking cyclooxygenase-1 or cyclooxygenase-2. J Clin Invest. 2000; 105:469–478. [PubMed: 10683376]
30. Suzuki R, Kohno H, Sugie S, et al. Strain differences in the susceptibility to azoxymethane and dextran sodium sulfate-induced colon carcinogenesis in mice. Carcinogenesis. 2006; 27:162–169. [PubMed: 16081511]
31. Clapper ML, Gary MA, Coudry RA, et al. 5-aminosalicylic acid inhibits colitisassociated colorectal dysplasias in the mouse model of azoxymethane/dextran sulfate sodium-induced colitis. Inflamm Bowel Dis. 2008; 14:1341–1347. [PubMed: 18452197]
32. Bissahoyo A, Pearsall RS, Hanlon K, et al. Azoxymethane is a genetic backgrounddependent colorectal tumor initiator and promoter in mice: effects of dose, route, and diet. Toxicol Sci. 2005; 88:340–345. [PubMed: 16150884]
33. Barrett CW, Fingleton B, Williams A, et al. MTGR1 Is Required for Tumorigenesis in the Murine AOM/DSS Colitis-Associated Carcinoma Model. Cancer Research. 2011; 71:1302–1312. [PubMed: 21303973] ** Very interesting study using MTGR1 deficient mice with AOM-DSS. This mice are resistant to CAC due to intra-tumoral apoptosis
34. Ashi KW, Inagaki T, Fujimoto Y, et al. Induction by degraded carrageenan of colorectal tumors in rats. Cancer Lett. 1978; 4:171–176. [PubMed: 647659]
35. Ishioka T, Kuwabara N, Oohashi Y, et al. Induction of colorectal tumors in rats by sulfated polysaccharides. Crit Rev Toxicol. 1987; 17:215–244. [PubMed: 2438086]
36. Oohashi Y, Ishioka T, Wakabayashi K, et al. A study on carcinogenesis induced by degraded carrageenan arising from squamous metaplasia of the rat colorectum. Cancer Lett. 1981; 14:267–272. [PubMed: 7332904]
37. Kohno H, Suzuki R, Sugie S, et al. Beta-Catenin mutations in a mouse model of inflammation-related colon carcinogenesis induced by 1,2-dimethylhydrazine and dextran sodium sulfate. Cancer Sci. 2005; 96:69–76. [PubMed: 15723650]
38. Seril DN, Liao J, Ho KL, et al. Dietary iron supplementation enhances DSS-induced colitis and associated colorectal carcinoma development in mice. Dig Dis Sci. 2002; 47:1266–1278. [PubMed: 12064801]
39. Hasegawa R, Sano M, Tamano S, et al. Dose-dependence of 2-amino-1-methyl-6-phenylimidazo[4,5-b]-pyridine (PhIP) carcinogenicity in rats. Carcinogenesis. 1993; 14:2553–2557. [PubMed: 8269626]
40. Doi K, Wanibuchi H, Salim EI, et al. Lack of large intestinal carcinogenicity of 2- amino-1-methyl-6-phenylimidazo[4,5-b]pyridine at low doses in rats initiated with azoxymethane. Int J Cancer. 2005; 115:870–878. [PubMed: 15751028]
41. Trivedi S, Wiber SC, El-Zimaity HM, et al. Glucagon-like peptide-2 increases dysplasia in rodent models of colon cancer. Am J Physiol Gastrointest Liver Physiol. 2012
42. Cooper HS, Everley L, Chang W, et al. The role of mutant Apc in the development of dysplasia and cancer in the mouse model of dextran sulfate sodium-induced colitis. Gastroenterology. 2001; 121:1407–1416. [PubMed: 11729120]
43. Tanaka T, Kohno H, Suzuki R, et al. Dextran sodium sulfate strongly promotes colorectal carcinogenesis in Apc(Min/+) mice: inflammatory stimuli by dextran sodium sulfate results in development of multiple colonic neoplasms. Int J Cancer. 2006; 118:25–34. [PubMed: 16049979]
44. Fujii S, Fujimori T, Kawamata H, et al. Development of colonic neoplasia in p53 deficient mice with experimental colitis induced by dextran sulphate sodium. Gut. 2004; 53:710–716. [PubMed: 15082590]

45. Chang WC, Coudry RA, Clapper ML, et al. Loss of p53 enhances the induction of colitis-associated neoplasia by dextran sulfate sodium. Carcinogenesis. 2007; 28:2375–2381. [PubMed: 17557903]
46. Shattuck-Brandt RL, Varilek GW, Radhika A, et al. Cyclooxygenase 2 expression is increased in the stroma of colon carcinomas from IL-10(−/−) mice. Gastroenterology. 2000; 118:337–345. [PubMed: 10648462]
47. Berg DJ, Davidson N, Kuhn R, et al. Enterocolitis and colon cancer in interleukin-10-deficient mice are associated with aberrant cytokine production and CD4(+) TH1- like responses. J Clin Invest. 1996; 98:1010–1020. [PubMed: 8770874]
48. Leach MW, Davidson NJ, Fort MM, et al. The role of IL-10 in inflammatory bowel disease: "of mice and men". Toxicol Pathol. 1999; 27:123–133. [PubMed: 10367687]
49. Kuhn R, Lohler J, Rennick D, et al. Interleukin-10-deficient mice develop chronic enterocolitis. Cell. 1993; 75:263–274. [PubMed: 8402911]
50. Normand S, Delanoye-Crespin A, Bressenot A, et al. Nod-like receptor pyrin domaincontaining protein 6 (NLRP6) controls epithelial self-renewal and colorectal carcinogenesis upon injury. Proceedings of the National Academy of Sciences of the United States of America. 2011; 108:9601–9606. [PubMed: 21593405]
51. Nguyen HTT, Dalmasso G, Torkvist L, et al. CD98 expression modulates intestinal homeostasis, inflammation, and colitis-associated cancer in mice. Journal of Clinical Investigation. 2011; 121:1733–1747. [PubMed: 21490400] ** Study focusing on the role of CD98 and cytokines in an AOM/DSS murine model
52. Yoshimi K, Tanaka T, Takizawa A, et al. Enhanced colitis-associated colon carcinogenesis in a novel Apc mutant rat. Cancer Science. 2009; 100:2022–2027. [PubMed: 19694754]
53. Wu S, Rhee KJ, Albesiano E, et al. A human colonic commensal promotes colon tumorigenesis via activation of T helper type 17 T cell responses. Nat Med. 2009; 15:1016–1022. [PubMed: 19701202]
54. Edwards RA, Witherspoon M, Wang K, et al. Epigenetic repression of DNA mismatch repair by inflammation and hypoxia in inflammatory bowel disease-associated colorectal cancer. Cancer Res. 2009; 69:6423–6429. [PubMed: 19638594]
55. Westbrook AM, Wei B, Braun J, et al. Intestinal mucosal inflammation leads to systemic genotoxicity in mice. Cancer Res. 2009; 69:4827–4834. [PubMed: 19487293]
56. Khor TO, Huang MT, Kwon KH, et al. Nrf2-deficient mice have an increased susceptibility to dextran sulfate sodium-induced colitis. Cancer Res. 2006; 66:11580–11584. [PubMed: 17178849]
57. Khor TO, Huang MT, Prawan A, et al. Increased susceptibility of Nrf2 knockout mice to colitis-associated colorectal cancer. Cancer prevention research (Philadelphia, Pa). 2008; 1:187–191.
58. Fukata M, Shang L, Santaolalla R, et al. Constitutive activation of epithelial TLR4 augments inflammatory responses to mucosal injury and drives colitis-associated tumorigenesis. Inflamm Bowel Dis. 2011; 17:1464–1473. [PubMed: 21674704]
59. Salcedo R, Worschech A, Cardone M, et al. MyD88-mediated signaling prevents development of adenocarcinomas of the colon: role of interleukin 18. The Journal of Experimental Medicine. 2010; 207:1625–1636. [PubMed: 20624890] * This paper studies how MyD88 decifient mice treated with AOM/DSS have an impaired epithelial repair
60. Lowe EL, Crother TR, Rabizadeh S, et al. Toll-like receptor 2 signaling protects mice from tumor development in a mouse model of colitis-induced cancer. PLoS One. 2010; 5:e13027. [PubMed: 20885960] * This study shows that TLR2 protects from CAC in an AOM/DSS mouse model
61. Xiao H, Gulen MF, Qin J, et al. The Toll-interleukin-1 receptor member SIGIRR regulates colonic epithelial homeostasis, inflammation, and tumorigenesis. Immunity. 2007; 26:461–475. [PubMed: 17398123]
62. Garlanda C, Riva F, Veliz T, et al. Increased susceptibility to colitis-associated cancer of mice lacking TIR8, an inhibitory member of the interleukin-1 receptor family. Cancer Res. 2007; 67:6017–6021. [PubMed: 17616656]
63. Rizzo A, Waldner MJ, Stolfi C, et al. Smad7 expression in T cells prevents colitisassociated cancer. Cancer Res. 2011; 71:7423–7432. [PubMed: 22028324]

64. Chiba T, Seno H, Marusawa H, et al. Host factors are important in determining clinical outcomes of Helicobacter pylori infection. J Gastroenterol. 2006; 41:1–9. [PubMed: 16501851]
65. Takai A, Marusawa H, Minaki Y, et al. Targeting activation-induced cytidine deaminase prevents colon cancer development despite persistent colonic inflammation. Oncogene. 2011
66. Garrett WS, Gallini CA, Yatsunenko T, et al. Enterobacteriaceae Act in Concert with the Gut Microbiota to Induce Spontaneous and Maternally Transmitted Colitis. Cell host & microbe. 2010; 8:292–300. [PubMed: 20833380] * This paper demonstrates the role of gut microbiota in TRUC mice induced CAC in the presence of *Enterobacteria*
67. Moser AR, Pitot HC, Dove WF. A dominant mutation that predisposes to multiple intestinal neoplasia in the mouse. Science. 1990; 247:322–324. [PubMed: 2296722]
68. Ritland SR, Gendler SJ. Chemoprevention of intestinal adenomas in the ApcMin mouse by piroxicam: kinetics, strain effects and resistance to chemosuppression. Carcinogenesis. 1999; 20:51–58. [PubMed: 9934849]
69. Reuter BK, Zhang XJ, Miller MJ. Therapeutic utility of aspirin in the ApcMin/+ murine model of colon carcinogenesis. BMC Cancer. 2002; 2:19. [PubMed: 12171603]
70. Chae WJ, Gibson TF, Zelterman D, et al. Ablation of IL-17A abrogates progression of spontaneous intestinal tumorigenesis. Proc Natl Acad Sci U S A. 2010; 107:5540–5544. [PubMed: 20212110]
71. Rudolph U, Finegold MJ, Rich SS, et al. Ulcerative colitis and adenocarcinoma of the colon in G alpha i2-deficient mice. Nat Genet. 1995; 10:143–150. [PubMed: 7663509]
72. Su LK, Kinzler KW, Vogelstein B, et al. Multiple intestinal neoplasia caused by a mutation in the murine homolog of the APC gene. Science. 1992; 256:668–670. [PubMed: 1350108]
73. Osburn WO, Kensler TW. Nrf2 signaling: An adaptive response pathway for protection against environmental toxic insults. Mutation Research - Reviews in Mutation Research. 2008; 659:31–39. [PubMed: 18164232]
74. Saleh M, Trinchieri G. Innate immune mechanisms of colitis and colitis-associated colorectal cancer. Nat Rev Immunol. 2011; 11:9–20. [PubMed: 21151034]
75. Fagarasan S, Kawamoto S, Kanagawa O, et al. Adaptive Immune Regulation in the Gut: T Cell-Dependent and T Cell-Independent IgA Synthesis. Annual Review of Immunology. 2010; 28:243–273.
76. Wang T, Cai G, Qiu Y, et al. Structural segregation of gut microbiota between colorectal cancer patients and healthy volunteers. ISME J. 2012; 6:320–329. [PubMed: 21850056]
77. Marchesi JR, Dutilh BE, Hall N, et al. Towards the Human Colorectal Cancer Microbiome. PLoS ONE. 2011; 6:e20447. [PubMed: 21647227]
78. Sobhani I, Tap J, Roudot-Thoraval F, et al. Microbial Dysbiosis in Colorectal Cancer (CRC) Patients. PLoS ONE. 2011; 6:e16393. [PubMed: 21297998]
79. Hoffmann C, Hill DA, Minkah N, et al. Community-Wide Response of the Gut Microbiota to Enteropathogenic Citrobacter rodentium Infection Revealed by Deep Sequencing. Infection and Immunity. 2009; 77:4668–4678. [PubMed: 19635824]
80. Fox JG, Ge Z, Whary MT, et al. Helicobacter hepaticus infection in mice: models for understanding lower bowel inflammation and cancer. Mucosal Immunol. 2011; 4:22–30. [PubMed: 20944559]
81. Tjalsma, H.; Boleij, A.; Kato, I. Streptococcus bovis and Colorectal Cancer Bacteria and Cancer. Khan, AA., editor. Springer Netherlands; 2012. p. 61-78.
82. Uronis JM, Mühlbauer M, Herfarth HH, et al. Modulation of the Intestinal Microbiota Alters Colitis-Associated Colorectal Cancer Susceptibility. PLoS ONE. 2009; 4:e6026. [PubMed: 19551144]
83. Wu S, Rhee K-J, Albesiano E, et al. A human colonic commensal promotes colon tumorigenesis via activation of T helper type 17 T cell responses. Nat Med. 2009; 15:1016–1022. [PubMed: 19701202]
84. Newman JV, Kosaka T, Sheppard BJ, et al. Bacterial Infection Promotes Colon Tumorigenesis in Apc Min/+ Mice. Journal of Infectious Diseases. 2001; 184:227–230. [PubMed: 11424022]
85. Chu F-F, Esworthy RS, Chu PG, et al. Bacteria-Induced Intestinal Cancer in Mice with Disrupted Gpx1 and Gpx2 Genes. Cancer Research. 2004; 64:962–968. [PubMed: 14871826]

86. Kado S, Uchida K, Funabashi H, et al. Intestinal Microflora Are Necessary for Development of Spontaneous Adenocarcinoma of the Large Intestine in T-Cell Receptor β Chain and p53 Double-Knockout Mice. Cancer Research. 2001; 61:2395–2398. [PubMed: 11289103]

87. Kim SC, Tonkonogy SL, Karrasch T, et al. Dual-association of gnotobiotic Il-10−/− mice with 2 nonpathogenic commensal bacteria induces aggressive pancolitis. Inflammatory Bowel Diseases. 2007; 13:1457–1466. [PubMed: 17763473]

88. Kim SC, Tonkonogy SL, Albright CA, et al. Variable phenotypes of enterocolitis in interleukin 10-deficient mice monoassociated with two different commensal bacteria. Gastroenterology. 2005; 128:891–906. [PubMed: 15825073]

89. Balish E, Warner T. Enterococcus faecalis Induces Inflammatory Bowel Disease in Interleukin-10 Knockout Mice. The American Journal of Pathology. 2002; 160:2253–2257. [PubMed: 12057927]

90. Erdman SE, Sohn JJ, Rao VP, et al. CD4+CD25+ Regulatory Lymphocytes Induce Regression of Intestinal Tumors in ApcMin/+ Mice. Cancer Research. 2005; 65:3998–4004. [PubMed: 15899788]

91. Erdman SE, Poutahidis T, Tomczak M, et al. CD4+ CD25+ Regulatory T Lymphocytes Inhibit Microbially Induced Colon Cancer in Rag2-Deficient Mice. The American Journal of Pathology. 2003; 162:691–702. [PubMed: 12547727]

92. Erdman SE, Poutahidis T. Roles for Inflammation and Regulatory T Cells in Colon Cancer. Toxicologic Pathology. 2010; 38:76–87. [PubMed: 20019355]

93. Kim SW, Kim HM, Yang KM, et al. Bifidobacterium lactis inhibits NF-κB in intestinal epithelial cells and prevents acute colitis and colitis-associated colon cancer in mice. Inflammatory Bowel Diseases. 2010; 16:1514–1525. [PubMed: 20310012] ** Great study from Korea investigating the role of Bifidobacterium lactis in an murine colitis model

94. Fukata M, Michelsen KS, Eri R, et al. Toll-like receptor-4 is required for intestinal response to epithelial injury and limiting bacterial translocation in a murine model of acute colitis. American Journal of Physiology - Gastrointestinal and Liver Physiology. 2005; 288:G1055–G1065. [PubMed: 15826931]

Key points

- Human colitis-associated cancers (CAC) are heterogeneous and can be induced by multiple oncogenic pathways. Thus, animal models are used in research to model this disease.
- The most frequently used CAC model induced by chemicals is the AOM-DSS model.
- Some genetic models such as $APC^{Min/+}$ and Giα2 knockout mice show a significant increase of intestinal tumors when they are under an inflammatory insult.
- Certain pathogenic bacteria can also promote tumorigenesis.
- Having a deficiency or over-expressing some innate immune key molecules also can promote the development of CAC.

Table 1

Overview of Genetic Models for Colitis-Associated Cancer.

Models of CAC	Result	Reference
NLRP6$^{-/-}$	increased AOM-DSS tumors	50
CD98 epithelial overexpression	increases AOM-DSS tumors; conditional epithelial KO protects	51
APC mutation +/− DSS or AOM or Bacteriodes fragilis infection	increases colonic tumors	5253
Giα2$^{-/-}$ spontaneous tumors	decreased MMR expression	54, 55
Nrf2$^{-/-}$	increased AOM-DSS tumors	56, 57
Villin-TLR4 (overexpressing TLR4)	increased AOM-DSS tumors	58
MyD88$^{-/-}$ or TLR2−/−	increased AOM-DSS tumors	59, 60
Single immunoglobulin IL-1-related receptor (SIGIRR)$^{-/-}$	increased AOM-DSS tumors	61
TIR8$^{-/-}$	increased AOM-DSS tumors	62
Smad7 T cell overexpression	protects against AOM-DSS tumors	63
Activation-induced cytidine deaminase (AID)$^{-/-}$	protects from IL-10$^{-/-}$ CAC	64, 65
TRUC mice (Tbet$^{-/-}$×Rag2$^{-/-}$) + *Klebsiella pneumonia* and *Proteus mirabilis*		66

Abbreviations: Nucleotide-binding oligomerization domain Leucine rich Repeat and Pyrin domain containing (NLRP), azoxymethane (AOM), dextran sodium sulfate (DSS), adenomatous polyposis coli (APC), mismatch repair (MMR), toll-like receptor 4 (TLR4), colitisassociated cancer (CAC).